Chapter 1: Methodology for Deriving U.S. EPA's Proposed Criteria for Lakes

This section describes the background, methodology, and results of U.S. EPA's analysis for deriving the proposed nutrient criteria for Florida lakes. The methodology includes developing lake classification and deriving chlorophyll and nutrient criteria for each of the resultant lake classes. All statistical data analysis was based on Florida's STORET and GWIS database, as developed and screened by Florida Department of Environmental Protection (FDEP).

1a. Methodology for Proposed Lake Classification

i. Background

Previous work on Florida lakes identified four lake classes: (1) soft water (acidic)-clear lakes; (2) acidic-colored lakes; (3) alkaline-clear lakes; and (4) alkaline-colored lakes. That classification system was originally proposed by Shannon and Brezonik (1972) on the basis of cluster analysis of lakes in north and central Florida. The classification was subsequently confirmed by Gerritsen et al. (2000) using principal components analysis of a much larger statewide data set compiled by Griffith et al. (1997). Among the lake groups, softwater (acidic) clear lakes of northwestern and central sandhills have been identified as extremely oligotrophic (Canfield et al. 1983).

Water color is due to dissolved organic carbon from decaying plant material in forests and wetlands of a lake's surface watershed. Color in Florida lakes ranges from none (clear) to heavily stained, tea-colored water. Color limits light penetration into the water column, and chlorophyll and algae are limited in heavily colored lakes. FDEP has defined colored lakes as those with color greater than 40 platinum-cobalt units (PCU). The color classification was based on the earlier analyses (e.g., Shannon and Brezonik 1972; Gerritsen et al. 2000; Paul and Gerritsen 2002).

Total alkalinity is a measure of the total concentration of bases in water expressed as calcium carbonate ($CaCO_3$) in milligrams per liter (mg/L). Those bases are usually bicarbonate (HCO_3^-) and carbonate ($CO_3^=$) that act as buffers that prevent drastic changes in pH. According to a survey of 946 Florida lakes, the total alkalinity ranges from 0.24 to 552 mg/L with a median value of 40 mg/L (Lazzarino et al. 2009).

Alkalinity of Florida lakes is regulated by the contribution of groundwater that has been in contact with limestone or calcareous soils (Stauffer and Canfield 1992). For example, acidic sandhill lakes often have no inlet or outlet but receive shallow groundwater from siliceous sand soils, and the shallow groundwater is perched above deeper limestone aquifers (Stauffer and Canfield 1992). Other lakes are not isolated from limestone aquifers, and groundwater contributes alkalinity from calcium carbonate dissolved from limestone. Alkalinity of Florida clear lakes ranges from zero (acidic lakes with no ability to neutralize acid) to well over 200 mg/L $CaCO_3$.

Alkaline lakes are considered to be more productive than acidic lakes, and alkalinity is associated with productivity of undisturbed reference lakes (Ryder et al. 1974; Oglesby 1977; Vighi and Chiaudani 1985). The effect of alkalinity has also been shown in experimental studies; increasing alkalinity from 13 to > 50 mg/L $CaCO_3$ increased primary productivity for the same nutrient loading (Arce and Boyd 1975). Alkalinity influences the species richness and composition of both aquatic macrophytes and algae species (Vestergaard and Sand-Jensen 2000). Alkalinity has also been used to classify lakes. For example, to comply with the European Water Framework Directive, Danish lakes were classified by depth and alkalinity; the alkalinity threshold used was 10 mg/L $CaCO_3$ (Søndergaard et al. 2005).

On the basis of recommendations from its Technical Advisory Committee (TAC), FDEP classified clear lakes on the basis of total alkalinity greater than or less than 50 mg/L (as $CaCO_3$). Because water color limits algal productivity, the TAC did not recommend an alkalinity breakdown in colored lakes. The TAC recommended that the clear lakes needed to be subcategorized to capture the differences between lakes receiving groundwater input from calcareous aquifer sources (higher alkalinity), which contain natural higher levels of phosphorus from lakes that receive most of their water from (low specific conductance) rainfall.

In many cases, historic alkalinity measurements did not exist to classify a clear lake as either alkaline or acidic. However, Florida has historically collected specific conductance measurements. When alkalinity data are unavailable, specific conductance can be used as a surrogate for alkalinity. Specific conductance is a measure of the ionic activity in water. A data analysis performed by FDEP and reexamined by U.S. EPA found that a specific conductance threshold value of 250 microsiemens/centimeter (μS/cm) corresponds to the alkalinity threshold of 50 mg/L (as $CaCO_3$) proposed by the TAC. Of those two measures, alkalinity is the preferred parameter to measure because it is less variable, and therefore a more reliable indicator, and also because it is a more direct measure of the presence of underlying geology associated with elevated nutrient levels.

ii. Data Used for Analysis

Water chemistry and chl. *a* data in Florida lakes were queried from the Florida STORET and the GWIS database. The initial data set consisted of 33,622 samples from 4,417 sites distributed within 1,599 lakes. All data were spatially linked to USGS lake reach codes on the basis of station coordinates. Water chemistry and chl. *a* data were averaged by parameter, lake reach code, and date. Data from 324 lakes were used to assess chl. *a* response to nutrients from more than 9,600 paired results. Nutrient concentrations, chlorophyll concentration, specific conductance, and alkalinity were log-transformed (natural log) for statistical analyses.

iii. Data Analysis for Classification

FDEP categorized lakes into clear and colored lakes on the basis of the geometric mean color for the period of record. A small number of lakes shifted between color categories over the period. Clear lakes were further subdivided on the basis of low alkalinity, or acidic (≤ 50 mg/L $CaCO_3$), and high alkalinity, or alkaline (> 50 mg/L $CaCO_3$), or specific conductance (using 250 μS/cm as the threshold). Where appropriate, data were log-transformed. Comprehensive nutrient, specific conductance, and chl. *a* data were available for 228 lake years in 78 clear lakes, and data including alkalinity were available for 200 lake years in 89 lakes.

iv. Other Classification Variables

Regional differences among the lakes were evaluated according to FDEP's stream nutrient regions (see Chapter 5 in USEPA 2009). Lakes showed similar chl. *a* responses regardless of location, with some differences in the range of nutrient concentrations (Figures 1-1 and 1-2). Therefore, regional difference were not found to be a significant differentiator of the relationship of total nitrogen (TN) or total phosphorus (TP) with chl. *a*. Figure 1-3 shows that there are no significant seasonal chl. *a* differences in the data set used to develop the nutrient criteria, indicating that a yearly average is appropriate.

v. Color and Alkalinity/Specific Conductance Results

FDEP has proposed an alkalinity class threshold at 50 mg/L. The justification for classifying lakes using alkalinity is that alkaline lakes appear to have higher natural nutrient concentrations than acidic lakes. If alkalinity differences exist among lakes but no related differences are in trophic variables or response, there is no reason for classifying lakes using alkalinity.

U.S. EPA examined regression relationships between chl. *a* concentration and TN and TP by FDEP-proposed lake categories (USEPA 2009). Chl. *a* concentrations exhibited statistically significant positive responses to both TP and TN on an annual average basis (e.g., Figure 1-4). On the basis of that analysis, clear lakes in general have lower nutrient concentrations than colored lakes and also have higher (50% to 100%) chl. a concentrations at the same TP or TN concentrations (Figure 1-4). Those relationships explain a large portion of the annual average variability observed in chl. *a* concentrations ($R2$ = 0.68–0.77). Although the slope of the chlorophyll response to nutrients is very similar between clear and colored lakes, the difference in chl. *a* response is enough to justify keeping the color classes separate.

Similarly, clear, alkaline lakes displayed higher nutrient and chl. *a* concentrations than clear, acidic lakes (Figure 1-5). Clear, acidic lakes have long been identified as crystal-clear, highly oligotrophic lakes (e.g., Canfield et al. 1983), and the TAC also recommended their separation as a class of lakes. Again, the slopes of the chlorophyll responses to TN and TP are similar between the alkalinity classes.

Accordingly, U.S. EPA concludes that three lake classes are appropriate, the same as recommended by FDEP and its TAC: clear lakes (≤ 40 PCU) that are acidic (≤ 50 mg/L $CaCO_3$ or specific conductance ≤ 250 µS/cm); clear lakes that are alkaline (> 50 mg/L $CaCO_3$ or specific conductance > 250 µS/cm); and colored lakes (> 40 PCU). U.S. EPA also recognizes that an alternative classification could be equally justified that uses a lower alkalinity/specific conductance threshold and possibly with an additional class division of acidic and alkaline colored lakes. For a description of further analyses of color and alkalinity, see Section 1f.ii.

1b. Methodology for Proposed Chlorophyll a Criteria

Because excess algal growth is associated with degradation in aquatic life and because chl. *a* levels are a measure of algal growth, U.S. EPA is using chl. *a* levels as indicators of healthy biological conditions, supportive of aquatic life in each of the categories of Florida's lakes described above. Multiple lines of evidence support chl. *a* criteria as an effective indicator of ambient conditions that would be protective of Florida's aquatic life use in lakes. Such lines of evidence include trophic state of lakes and aquatic life use, historical reference conditions in Florida lakes, and model results.

i. Trophic State and Aquatic Life Use

Trophic State

One part of recreational use protection is maintaining ecosystem integrity, especially with respect to some highly visible attributes of the system. In lakes, important visual attributes for users are water clarity and color. That does not mean that all lakes should be crystal-clear, rather, that the lakes should resemble their natural, expected conditions of color, clarity, and productivity. For waters that are exceptional or nearly pristine, they should very closely resemble all aspects of the natural condition.

Waters are typically classified into three trophic classes to reflect nutrient conditions and overall productivity: oligotrophic, mesotrophic, and eutrophic. Classification of a lake into one of the classes is commonly done with a Trophic State Index (TSI). The TSI quantifies the trophic state (oligotrophic, mesotrophic, eutrophic) in a waterbody on the basis of observed measurements of nutrients and chl. *a*. The trophic classes and the TSI are explained below and also in U.S. EPA 2000. For Florida, a TSI of 40–50 corresponds to the boundary between oligotrophic and mesotrophic, and a TSI of 60–70 corresponds to the boundary between mesotrophic and eutrophic conditions (Salas and Martino 1991; USEPA 2009). Anthropogenically caused eutrophic conditions are generally acknowledged to be undesirable for drinking water, recreation, and aquatic life uses. In the past, FDEP has managed for a maximum TSI of 60 (highest level for mesotrophy in Florida), which corresponds to chl. *a* concentration of 20 μg/L. As a primary line of evidence, U.S. EPA reviewed and evaluated TSI information from FDEP in deriving chl. *a* criteria that are protective of designated aquatic life uses in Florida's lakes.

FDEP has a long history of using a modification of Carlson's TSI (Carlson 1977) as a measure of lake trophic state and lake water quality for the state's 305(b) and 303(d) assessments. Trophic state reflects the biological response to several factors, including nutrient effects on phytoplankton chl. *a*, which can be modified or mitigated by water retention time, grazing, and macrophyte nutrient uptake. Havens (2000) reported that the TSI approach provides an effective, low-cost method for tracking long-term changes in pelagic structure and function and has value in monitoring lake ecology and responses to management actions.

Carlson's original TSI classified lakes on the basis of chl. *a* levels and nitrogen and phosphorus concentrations, and it included three indicators—secchi depth, chl. *a*, and TP—to describe a lake's trophic state. A 10-unit change in the index represents a doubling or halving of secchi transparency, TP, and algal biomass (but note that chl. *a* does not follow the doubling or halving precisely; Carlson 1977).

The following interpretation scheme for the TSI is based on nutrient/chl. *a* responses in north temperate lakes (Carlson and Simpson 1996):

TSI < 30	Classical Oligotrophy: Clear water, oxygen throughout the year in the hypolimnion, salmonid fisheries in deep lakes.
TSI 30–40	Deeper lakes still exhibit classical oligotrophy, but some shallower lakes will become anoxic in the hypolimnion during the summer.
TSI 40–50	Water moderately clear but increasing probability of anoxia in hypolimnion during summer.
TSI 50–60	Lower boundary of classical eutrophy: Potential for decreased transparency, anoxic hypolimnia during the summer and macrophyte growth, warm-water fisheries only.
TSI 60–70	Dominance of blue-green algae, algal scums probable, extensive macrophyte problems.
TSI 70–80	Heavy algal blooms possible throughout the summer, dense macrophyte beds but extent limited by light penetration. Often would be classified as hypereutrophic.
TSI > 80	Algal scums, summer fish kills, few macrophytes, dominance of rough fish.

Salas and Martino (1991) proposed an alternative TSI categorization based on their work in phosphorus-limited, warm-water tropical lakes, which is more directly applicable to Florida conditions. The TSI and chl. *a* values in Table 1-1 were determined on the basis of the TSI relationship with TP. In the original index, a TSI of 50–60 represents the lower boundary of eutrophy in temperate lakes, Salas and Martino considered that same range of TSI values to be mesotrophic in warm-water lakes.

The TSI equation describes a theoretical relationship between chl. *a*, TP, and TN. In the revised TSI, chl. *a* doubles with every 10-point increase in the TSI (Table 1-2).

As part of the state's 305(b) assessment, FDEP revised the TSI by (a) replacing secchi depth with TN, and (b) adding equations that adjust the nutrient component of the TSI to reflect the limiting nutrient. Use of secchi depth in Florida as a contributing measure to TSI is limited because of the high frequency of dark-water lakes (> 40 PCU), where tannins, rather than algae, diminish transparency.

FDEP's resultant TSI is based on chl. *a*, TN, and TP concentrations, as follows:

$$TSI = (CHLATSI + NUTRTSI) / 2 \qquad [1]$$

Where,

$$CHLATSI = 16.8 + 14.4 \times \mathrm{LN}\ (\mathrm{CHLA}) \qquad [2]$$

NUTRTSI is based on limiting nutrient considerations, as follows:

If TN / TP > 30, then lake is considered phosphorus limited and

$$\mathrm{NUTRTSI} = \mathrm{TP2TSI} \qquad [3]$$

$$\mathrm{TP2TSI} = 10 \times [2.36 \times \mathrm{LN}(\mathrm{TP} \times 1{,}000) - 2.38] \qquad [4]$$

If TN / TP < 10, then lake is considered nitrogen limited and

$$\mathrm{NUTRTSI} = \mathrm{TN2TSI} \qquad [5]$$

$$\mathrm{TN2TSI} = 10 \times [5.96 + 2.15 \times \mathrm{LN}(\mathrm{TN} + 0.0001)] \qquad [6]$$

If 10 < TN / TP < 30, then lake is co-limited and

$$NUTRTSI = (TPTSI + TNTSI)/2 \quad [7]$$
$$TNTSI = 56 + [19.8 \times LN(TN)] \quad [8]$$
$$TPTSI = [18.6 \times LN(TP \times 1000)] - 18.4 \quad [9]$$

Equations 1-9 were determined on the basis of the analysis of data from 313 Florida lakes and were adjusted so that a chl. *a* concentration of 20 µg/L was equal to a TSI value of 60. For the 1998 305(b) report, a TSI threshold of 60 was used to represent "fair" lakes, while lakes above 70 were assessed as "poor."

During development of the Impaired Waters Rule (IWR) in 1999–2000, the IWR TAC reviewed the TSI used in the 305(b) assessment and recommended that it be used to assess lakes for impairment. On the basis of then current U.S. EPA guidance that "fair" waters should be included on state 303(d) lists, the TAC recommended that the nutrient impairment threshold for most lakes (for those with a color higher than 40 PCU) should be an annual average TSI of 60.

While the TAC recommended use of the TSI threshold of 60 for most lakes, it also recognized that some lakes are naturally oligotrophic and have significantly lower natural background TSIs. The TAC requested that FDEP evaluate data from reference lakes from the FDEP's Bioassessment Sampling Program using principal components analysis in an attempt to identify different types of lakes on the basis of water quality with the goal to establish different TSI thresholds for each type.

While many different parameters were evaluated, the analysis initially focused on a four-part chemical classification of Florida lakes consisting of acid-clear, acid-colored, alkaline-clear, and alkaline-colored. That classification system had originally been proposed by Shannon and Brezonik (1972) and was subsequently confirmed as part of the development of the Lake Condition Index for Florida (Gerritsen et al. 2000). However, the analysis conducted for the IWR indicates that the most significant differences in the TSI and TSI-related parameters (nutrients and chlorophyll) were seen when the lakes were classified by color alone, with lakes with a color of less than 40 PCU having significantly lower TSIs. That color classification system also covered a previously identified target population of oligotrophic lakes that the TAC wanted to address (low color, oligotrophic lakes in the panhandle region of Florida). The FDEP then recommended, and the TAC agreed, to establish a TSI threshold of 40 for those lakes, which is equivalent to a chl. *a* of 5 µg/L.

Furthermore, the TAC recommended that clear, but higher specific conductance lakes should be held to a 20 µg/L chl. *a* threshold for aquatic life use protection. The higher threshold acknowledges the fact that the higher specific conductance lakes would be expected to have higher chl. *a* levels related to the higher natural phosphorus conditions.

Aquatic Life Use

Nearly all Florida lakes are shallow and, thus, include large areas of potential habitat for submerged and floating-leaved plants. If the water is relatively clear, shallow zones of the lakes can be dominated by native submerged and floating plants. Natural primary production in such shallow lakes includes contributions from both submerged macrophytes and phytoplankton. If the water is turbid from suspended sediment or excess algae, the submerged plants will not receive sufficient light to survive.

The algae and macrophytes in shallow lakes have complex dynamics under nutrient enrichment and physical disturbance conditions (Scheffer et al. 1993). Submerged macrophytes intercept nutrients and incorporate them into plant biomass, where the nutrients are then unavailable to the phytoplankton algae. Shallow, mesotrophic lakes with abundant macrophytes have clear water and relatively low chlorophyll concentrations. Such lakes are mesotrophic (intermediate level of productivity), but their water column chlorophyll concentrations are low because a large fraction of primary production is from the macrophytes and not phytoplankton algae. Such a stable state can be upset by (1) sudden loss of macrophytes because of disturbance (e.g., grass carp, hurricane, other disturbance); or (2) excessive nutrient loading leading to increased epiphytes and phytoplankton, which then shade the macrophytes (e.g., Scheffer et al. 1993, 2001; Bachmann et al. 1999, 2002; Lowe et al. 1999, 2001). Loss of the macrophytes can further increase TP concentrations (Bachmann et al. 2002), possibly from release from biomass and sediment, and further increasing TP available to the phytoplankton. The new condition can also be stable, and turbidity from algae or resuspension of bottom sediment prevents macrophytes from reestablishing. Excess plant biomass problems can occur both ways: dense algal blooms or a lake choked with invasive floating plants (e.g., *Hydrilla*, *Myriophyllum*, water hyacinth); extremely dense floating plants can also be in a stable state (Scheffer et al. 2003). Substantial evidence from both historical records and paleolimnology shows that several Florida lakes have made the transition from macrophyte-dominated, mesotrophic clear waters to phytoplankton-dominated, eutrophic (or hypereutrophic) turbid waters as the consequence of nutrient enrichment (Lowe et al. 1999, 2001; Kenney et al. 2002).

Submerged vegetation and phytoplankton support different consumer food webs. Macrophytes are the origin of a detritus- and periphyton-based food web including macroinvertebrates, amphibians, fish, and birds. The phytoplankton food web supports zooplankton and fish. More species of fish are dependent on littoral, benthic macroinvertebrates for their diet than those that are dependent on zooplankton (e.g., Vadeboncoeur et al. 2002). In addition to supporting a food web, macrophytes also provide habitat and cover for fish (especially smaller forage fish and juvenile fish) and larger macroinvertebrates (e.g., Wetzel 1975). Vegetated areas of lakes have greater animal diversity (invertebrates, fishes, birds) than open waters (Havens et al. 1996). Collapse of the submersed vegetation results in an ecosystem change—the overall simplification of food webs and system function; and reduction or elimination of native fish, invertebrate, and bird species dependent on the littoral habitat. Such effects are well-documented from eutrophication effects in the Great Lakes and elsewhere (e.g., Wetzel 1975; Winfield 2004). Thus, protecting aquatic life use requires maintenance of these system functions.

Because submersed vegetation is dependent on light, maintaining a lake's historic balance between algae and submerged plants requires maintaining overall historic water transparency. Natural transparency varies widely in Florida lakes because of the range in water color from crystal-clear to deeply stained. Regardless of what the natural transparency of a lake might be, it is reduced by increased algal growth that results from anthropogenic nutrient enrichment. Carlson's TSI incorporates an empirical relationship between secchi transparency and chlorophyll, such that an increase in chlorophyll concentration by a factor of approximately 2.75 corresponds to a 50% decrease in secchi transparency (Carlson 1977). For constant water color, that relationship is expected to hold in Florida lakes. Accordingly, maintaining water column chlorophyll concentrations within traditional oligotrophic or mesotrophic limits reduces the risk that submerged vegetation and system functions will be unacceptably altered. U.S. EPA recognizes that total nutrient loading to a lake might be higher than the apparent trophic state measured in the water column because submerged macrophytes intercept some fraction of the nutrients.

ii. Reference Condition

The reference condition approach entails identifying lakes exhibiting minimally disturbed conditions, collecting and analyzing water quality data from the lakes, and calculating an upper percentile of the data distribution to establish the criteria. Such an approach assumes that minimally disturbed conditions are the target condition, and substantial departure from this condition, as expressed by the percentile, should be prevented. Several approaches can be used to identify a reference condition: identifying specific reference lakes; paleolimnological investigation to determine historic and prehistoric conditions; statistical or theoretical models to infer reference conditions when they are not available everywhere.

Identification of minimally disturbed lakes (reference approach)

FDEP has identified reference lakes throughout the state, defined as lakes with the least amount of land use change (mostly natural land cover) within 500 meters of the lakeshore. An earlier report (Paul and Gerritsen 2002) characterized chl. *a* in five classes of reference lakes, and more recently, FDEP analyzed the distribution of chl. *a* in reference colored and clear lakes (but did not further break down the analysis by alkalinity/specific conductance classes, see Appendix A-1).

A reference site approach, coupled with other techniques, including contour plot interpolation, was suggested as a method to establish chl. *a* thresholds in Florida (Paul and Gerritsen 2003). On the basis of the 75th percentile of reference sites, determined by contour plot interpolation, Paul and Gerritsen (2003) proposed chl. *a* targets for Florida clear lakes (< 40 PCU) ranging from 2 μg/L to 8 μg/L, and colored lake targets ranging from 9 μg/L to 18 μg/L.

Paleolimnological Studies

Another line of evidence that supports U.S. EPA's proposed chl. *a* criteria is historical reference conditions as inferred from paleolimnological studies. Diatom frustules can be preserved in lake sediments, and if the sediments are undisturbed can be used to infer historic and prehistoric conditions in the lake. Because species composition of diatoms is highly sensitive to water column nutrient concentrations, nutrient concentrations and pH can be inferred from the analysis.

Paleolimnological studies in Florida, where pre-human disturbance chl. *a* values were inferred from an analysis of diatom communities in deep sediment cores, show that historical conditions in clear, acidic panhandle lakes are similar to present-day reference conditions with TP concentrations in the range of 1 to 10 μg/L, and historical TP concentrations in alkaline peninsular lakes were in the range of 15–90 μg/L (mixture of both clear and colored lakes). The acidic panhandle lakes would be considered to be at the boundary between classical oligotrophy and mesotrophy.

The paleolimnological studies suggest that most peninsular Florida lakes would be considered to be at (or above) the lower boundary of classical eutrophy, even before human habitation of the state (Whitmore and Brenner 2002; Whitmore 2003). Paleolimnological studies conducted at (colored) Lakes Shipp, Lulu, Haines, May, Conine and Bonny in the Florida peninsula suggest that the average chl. *a* in the lakes would naturally range between 14–20 µg/L. This is one line of evidence for supporting the chl. *a* threshold of 20 µg/L that was part of the IWR's TSI threshold. However, paleolimnolgy of (colored) Lakes Wauberg and Hancock suggests that historic chl. *a* in those lakes naturally were in the ranges of 38–48 µg/L and 74–133 µg/L, respectively. Note that although Lake Hancock might be somewhat atypical, the paleolimnological results suggest that any proposed nutrient criteria will need to allow for site-specific alternative criteria (SSAC) in those lakes with naturally higher or lower nutrient levels.

Model Predictions

In addition to present-day reference and paleolimnological reconstruction, U.S. EPA also used information from a model developed by the St. Johns River Water Management District that predicts nutrient and chl. *a* concentrations for a lake given its depth, alkalinity, and color to support the proposed chl. *a* criteria.

The St. Johns River Water Management District developed a statistical model of reference condition (transparency and chlorophyll) using observations of existing reference lakes (Lowe et al., personal communication). The model is based on observations that the natural, reference fertility of a lake can be predicted from its depth and alkalinity (Ryder et al. 1974; Oglesby 1977; Vighi and Chiaudani 1985). This model is termed the Morphoedaphic Index, or MEI, model because it makes use of lake morphometry (depth), and edaphic factors (the effects of alkalinity from influence of deep groundwater). Development of the model is explained in Appendix A-2. Application to Florida lakes also required inclusion of water color. The model predicts natural, reference chlorophyll concentrations for any lake given its depth, alkalinity, and color. The model predicted chlorophyll concentrations for 10% reduction of water transparency for a set of lakes identified as clear versus colored and acidic vs. alkaline. Such predictions were in accordance with the 20 µg/L chl. *a* concentration limit defined by the TSI mesotrophic-eutrophic boundary and with 5–10 µg/L for the oligotrophic-mesotrophic boundary.

iii. Other Efforts to Establish Chlorophyll a Thresholds

Appendix A-3 contains a review of literature pertaining to establishing protective chl. *a* thresholds, predominantly from the United States. The literature suggests six main approaches for establishing protective chl. *a* thresholds in lakes:

- Using an upper percentile of the distribution of reference lakes (used by U.S. EPA; above)
- Paleolimnologic studies, where preindustrial disturbance chl. *a* values are inferred from an analysis of diatom communities in deep sediment cores (used by U.S. EPA; above)
- Expert elicitation for determining protective TSI or chl. *a* values (partially used by FDEP; below)
- Direct responses of other biological attributes (invertebrate community, vegetation, fish community) (evaluated but not used; below)
- Associating lake user visual perceptions (for swimming and aesthetics) with simultaneously measured chl. *a* (evaluated but not used; below)

- Setting the criterion to maintain the existing condition (maintenance strategy) (evaluated but not used; below).

Expert Opinion

Several states, including Florida, have used input from scientific advisory committees to establish chl. *a* or TSI targets. Recommendations of Florida's TAC are discussed in 1b.i, above. Virginia and Iowa queried a panel of experts to establish protective chl. *a* targets, and the scientists in both states independently arrived at a recommended level of 25 µg/L as a yearly average (Gregory 2007; Wilton 2008). Virginia's panel further recommended that chl. *a* not exceed 50 µg/L as an instantaneous measurement. Arizona, using a TSI and a weight-of-evidence approach, established lake summer time (peak season) chl. *a* targets at 20–30 µg/L (ADEQ 2008). Maryland established lake summer time chl. *a* targets using the rationale that a TSI of 50 (10 µg/L chl. *a*) would prevent mesotrophic lakes from becoming eutrophic and that a TSI of 60 (20 µg/L chl. *a*) would protect against excessive eutrophication (Rule 2004). West Virginia used expert opinion to establish a chl. *a* threshold of 33 µg/L. Additionally, Iowa has established annual average TMDL targets in specific lakes for the protection of aquatic life use (Lost Lake) using a chl. *a* threshold of 33 µg/L (USEPA 2008).

The various thresholds developed by expert opinion that would protect against excessive eutrophication, expressed as annual or summertime averages, yields a range of 20–33 µg/L of chl. *a*. That range of values suggests that Florida's IWR TAC recommendation of 20 µg/L in colored lakes is as protective as those established by many other states.

Biological Responses

The responses of valued ecological attributes, such as benthic macroinvertebrates, fish, or submerged macrophytes, to various chl. *a* levels, would provide the most direct method for establishing targets that would protect aquatic life. FDEP established a Lake Condition Index, using benthic macroinvertebrates as a response variable in lakes (Gerritsen et al. 2000), and a Lake Vegetation Index, using the littoral and shorezone macrophyte community as the response indicator (Fore et al. 2007). Although initial results were promising, FDEP eventually concluded that color was more responsible for explaining benthic response than were human disturbance measures, such as the Landscape Development Intensity Index (Fore 2007). Development and use of the Lake Vegetation Index is explained in Appendices A-4 and A-5.

Other states have used a fisheries response variable. For example, Virginia conducted an analysis to determine the effect of chl. *a* levels on the health of fisheries and concluded that summer average chl. *a* concentrations of 25 µg/L in cool-water lakes and 35–60 µg/L in warm-water lakes were protective of fish health (Gregory 2007). Minnesota—using multiple lines of evidence, including regional patterns, reference lakes, fish response, lake user perception, paleolimnolgy, and nuisance algal bloom frequency—established summer mean chl. *a* targets of 3 to 5 µg/L for designated coldwater trout fisheries, 9–22 µg/L for deep lakes, and 20–30 µg/L for shallow (< 4.5 m) lakes (Heiskary and Wilson 2008). Colorado proposed that summer average chl. *a* be maintained below 25 µg/L to assure high-quality fisheries (Saunders 2009).

FDEP is investigating fish community composition data collected from Florida lakes by the Florida Fish and Wildlife Conservation Commission (FWC) for comparison to chl. *a* data.

Results of initial analyses do not yield a notable response signal in the data and, thus, do not further inform the determination of chl. *a* targets.

FDEP has developed a preliminary Lake Vegetation Index, which comprises all submerged, floating, emergent, and woody vegetation in a lake's littoral and shore zones. The index was not developed or calibrated for response to nutrients and, thus, responds primarily to general levels of anthropogenic disturbance and less to nutrient loading. The general response of submerged aquatic macrophytes to eutrophication is well established and is described above in section 1b.i. However, quantitative information is not yet available on submerged macrophytes in Florida to examine potential thresholds of response.

User Perceptions

In Texas, a study of lake user perceptions indicated that in reservoirs without inorganic turbidity (> 1 m secchi) chl. *a* levels below approximately 20–25 µg/L still support full immersion recreational uses, as well as aesthetics (Glass 2006). For this study, lake users were asked to fill out a questionnaire concerning their visual perceptions (for swimming and aesthetics) while chl. *a* was simultaneously measured. A similar study conducted in Florida demonstrated that user perceptions differed depending on the lake region (Hoyer et al. 2004). In the Florida study, when lake users responded to a question concerning suitability of the lake for recreation and aesthetic enjoyment by saying, "beautiful, could not be nicer," chl. *a* ranged from approximately 30 µg/L in the Central Valley Lake region (generally high color) to approximately 3 µg/L in the Trail Ridge Region (generally uncolored lakes) (Hoyer et al. 2004). Those studies did not associate chl. *a* values with public health concerns, only the public perception of whether swimming was desirable. The user perceptions support the concept of different chl. *a* criteria for clear lakes and colored lakes.

Maintaining Existing Conditions

Alabama's approach to establishing lake or reservoir chl. *a* targets could be described as a method designed to "maintain the existing condition" (Macindoe 2006). Alabama's chl. *a* targets for specific lakes or reservoirs range from 5 µg/L to 27 µg/L. The Florida IWR TAC's recommendation of a TSI of 40 for Florida clear reference lakes was based on the concept of maintaining the current condition of panhandle region sandhill lakes. A TSI of 40 equates to a chl. *a* value of 5µg/L.

Relationships between Cyanobacteria Abundance and Chl. *a*

It is well established that cyanobacteria can become very abundant and completely dominate the phytoplankton community in lakes when conditions are favorable. Some cyanobacteria blooms can be toxic and present a health risk to people recreating in and on the water. The World Health Organization (WHO) has established recommendations for recreational exposure during cyanobacterial blooms (WHO 1999). Its recommendations reflect findings that a chl. *a* level of 10 µg/L in which cyanobacteria are dominant presents a relatively low probability of mild irritative or allergenic effects, while a chl. *a* level of 50 µg/L in which cyanobacteria are dominant presents a moderate risk of adverse health effects. The WHO guidance for recreation in waters (WHO 2003, section 8.1) states that 46 species of cyanobacteria have been shown to cause toxic effects in vertebrates and that any species or genus of cyanobacteria cannot be ruled out as potentially toxic. WHO cautions that, "it is prudent to presume a toxic potential in any cyanobacterial population."

Because of the potential human health risks associated with cyanobacteria blooms, FDEP considered the possibility of a chl. *a* threshold that might be associated with a high probability of cyanobacteria blooms. FDEP examined the relationship between chl. *a* and the percent cyanobacteria in 1,364 phytoplankton samples from small and large lakes randomly sampled between 2000 and 2006 in Florida's probabilistic sampling network. Figure 1-6 shows chl. *a* values regressed against the percent cyanobacteria for each sample. According to the graph, no apparent increased probability of cyanobacteria dominance exists as chl. *a* increases. Samples dominated by one of the 13 harmful algal bloom taxa listed by WHO (WHO 2003, section 8.1) do not show an increasing trend of cyanobacteria dominance with chl. *a* either. That analysis does not further inform the determination of chl. *a* targets.

Conclusions

Carlson and Simpson (1996) noted that trophic state is not synonymous with the concept of water quality. While trophic state is an absolute scale that describes the biological condition of a waterbody, water quality is used to describe the condition of a waterbody in relation to human needs or values, relative to the use of the water and the expectations of the user. Water quality standards are created to protect the designated uses of waterbodies. In Florida lakes, the designated uses are for the protection of healthy, well-balanced populations of fish and wildlife, and for recreation in and on the water. Criteria must provide protection for these sometimes competing interests. For example, an oligotrophic to mesotrophic lake could have water quality deemed desirable for swimming, as well as natural communities of fish, invertebrates, and submerged macrophytes. However this same lake might not be considered to be optimal for bass fishing. For that reason, U.S. EPA has taken a weight-of-evidence approach for establishing protective chl. *a* thresholds.

Multiple lines of evidence were also used to evaluate the rigor of protection inherent in the IWR TAC's TSI-based chl. *a* recommendations, which were adopted into the IWR in 2002 (Chapter 62-303, FAC). The multiple lines of evidence considered above, including aquatic life use protection, observed and modeled reference condition, and paleolimnology, converge to support the Florida IWR TAC's original recommendation.

Colored water lakes – On the basis of the above multiple lines of evidence, U.S. EPA proposes that an annual average chl. *a* of 20 μg/L in lakes with water color greater than 40 PCU is protective of designated uses.

Clear, acidic lakes – Some lakes are naturally oligotrophic and have significantly lower natural background nutrient and chl. *a* concentrations. Examples include the "sandhill" lakes in the Florida panhandle, which are clear and oligotrophic, often with chlorophyll as low as 1 μg/L. For protection of those and other clear, acidic lakes, U.S. EPA recommends a chl. *a* criterion of 6 μg/L. In contrast to colored lakes and clear, alkaline lakes, clear, acidic lakes do not receive significant natural nutrient inputs from groundwater or other surface water sources. The lakes are, thus, expected to be oligotrophic. Some of Florida's clear, acidic lakes, in sandhills in northwestern and central Florida, have been identified as extremely oligotrophic with chl. *a* levels of less than 2 µg/L (e.g., Canfield et al. 1983; Griffith et al. 1997). As discussed above, the warm-water TSI studies suggest that the oligotrophic-mesotrophic boundary occurs at chl. *a* concentrations of approximately 7 µg/L. FDEP's Nutrient Criteria TAC recommended a chl. *a* concentration of 9 µg/L. The TAC based its recommendation largely on an analysis of lake data that showed lack of association between an index of benthic macroinvertebrate health and chl. *a* in the range of 5–10 µg/L chl. *a*. Within such a small range at the oligotrophic-mesotrophic boundary, lack of association is not surprising because the stressor (chl. *a*) range is small and because the invertebrate indicator is responsive to numerous aspects of natural conditions and stressors.

However, some evidence of meaningful distinctions exists within the range based on endpoints more directly responsive to nutrients. In this case, the Florida MEI model predicted reference chl. *a* concentrations in the range of 1.4–7.0 µg/L (with seven of the eight values below 5 µg/L) for a set of clear, acidic lakes in Florida and predicted a 10% loss of transparency when chl. *a* concentrations in the range of 5.6–11.8 µg/L (with five of the eight values below 7 µg/L). All but one of the clear, acidic lakes had predicted natural or reference conditions below 6 µg/L chl. *a* and the clear majority (six of eight) of clear, alkaline lakes had predicted 10% transparency loss above 6 µg/L chl. *a.* Given the available information on reference condition and predicted effect levels, U.S. EPA adjusted the approximate oligotrophic-mesotrophic boundary value of 7 µg/L slightly downward to 6 µg/L as the proposed chl. *a* criterion.

Clear, alkaline lakes – Earlier, in 2002, FDEP's IWR TAC had recommended a chl. *a* criterion of 5 µg/L in clear lakes that was based on a "maintain existing condition approach" and that was primarily targeted at a specific geographic region of Florida (the panhandle). The TAC also suggested that different nutrient and chl. *a* expectations should be established for high specific conductance (> 100 µS/cm) clear lakes because of the naturally higher, aquifer-derived phosphorus levels in that subset of clear lakes. U.S. EPA proposes the TAC suggested nutrient thresholds in clear, high-specific conductance lakes be based on preventing the annual average chl. *a* from exceeding 20 µg/L.

1c. Methodology for Proposed TP and TN Criteria in Lakes

Paired nutrient and chl. *a* data were available for 129 colored lakes. Initial analyses revealed statistically significant ($p < 0.001$) yet weak relationships between chl. *a* and TP (*R2* = 0.38) and TN (*R2* = 0.47). Other factors influencing the chl. *a* response were then investigated in an attempt to improve the relationship of chl. *a* with nutrients. Despite initial lake subcategorization by color, a significant inverse relationship (Spearman *R* = –0.25) remained between color and chl. *a*, with the influence of color most pronounced in lakes with color in excess of 150 to 200 PCU. Chl. *a* level in these highly colored lakes were typically reduced when compared to the levels in less colored lakes, despite similar nutrient concentrations; that is, color in excess of approximately 150 PCU depresses the nutrient response (light limitation).

A multiple regression model (adjusted *R2* = 0.507) was constructed between chl. *a* (dependent variable) and TP and TN (independent variables) to investigate the influence of lake color on chl. *a* response in colored lakes (Table 1-3). Model residual error was plotted against both color expressed as a long-term geometric mean (period of record) and annual geometric mean color. This evaluation demonstrated that the nutrient regression model tended to underestimate (positive residuals) chl. *a* concentrations at lakes with color less than approximately 150 PCU and overestimated (negative residuals) chl. *a* levels at lake with color over approximately 150 PCU (Figure 1-7). Classification and Regression Tree (CART) analysis was used to discriminate a breakpoint in the model residual error. A significant breakpoint that explained 36.4% of the relative error in the residuals was found at a long-term lake color of 143 PCU. Additional breakpoints were found at annual geometric mean colors of 54 and 360 PCU. Those subsequent breakpoints provided only marginal improvements in the amount of explained variance (Figure 1-8). On the basis of the CART analysis, the colored lakes were further subcategorized to long-term ranges of > 40–140 PCU (moderately colored; n = 100 lakes) and > 140 PCU (highly colored; n = 29 lakes), for purposes of investigating nutrient responses to account for the substantial remaining influence of color on the chl. *a*.

Regional differences among the moderately colored lakes (color between 40 and 140 PCU) were evaluated, but U.S. EPA found that those colored lakes show similar chl. *a* responses regardless of location, although there were differences in the range of nutrient concentrations (Figures 1-9 and 1-10). Chl. *a* exhibited statistically significant positive responses to both TP and TN on an annual average basis in the moderately colored lakes (Figure 1-11). Such relationships are sufficiently robust to develop scientifically defensible and protective criteria.

The relationships between TP and TN and chl. *a* in the highly colored lakes (greater than 140 PCU) were significant but weak (Figure 1-12). The relationships demonstrate that nutrients influence chl. *a* response (excess algal growth) in highly colored lakes and thus provide support for the need to develop numeric nutrient criteria to protect the designated use. However, the relationships are not sufficiently robust to directly derive numeric nutrient criteria given the high level of uncertainty and unexplained variance. Without a strong and robust nutrient-chl. *a* relationship in the highly colored (> 140 PCU) lakes, fully protective criteria for these systems can be developed on the basis of the response relationships from the moderately colored lakes (40–140 PCU), although the criteria will be somewhat overprotective, given that high color will reduce algal response and biomass.

Regression models describe the relationship between two variables where the magnitude of one variable (dependent) is assumed to be a function of one or more independent variables; that is, a degree of the variance in the dependent variable is explained by the independent variable(s). The regression line and equation define the average relationship (i.e., half the data points fall above the line and half below it). Essentially, there is a 50% probability that a given level of nitrogen or phosphorus will elicit the chl. *a* response corresponding to the regression equations shown in Figures 1-11 and 1-13. FDEP concluded that a simple application of an average response was not adequately protective and that a more complex application was needed to account for uncertainty in the dose-response relationship.

For nutrient criteria, response uncertainty can be managed by considering nutrient concentrations in a range between a level that is unlikely (e.g., 25% probability) to elicit a given threshold of response and a level that that is likely to elicit a response (e.g., 75% probability). Regression prediction intervals provide a range above and below the regression line that incorporate the unexplained variability of the independent variable as well as the uncertainty in the model parameters (slope and intercept). Within the range of nutrient concentrations (between the upper and lower prediction interval), there is less certainty that a response (exceedance of the chlorophyll target) will or will not occur. This represents a range of conditions in which nutrients can be managed while considering the potential for Type I (incorrectly identifying a water as impaired) and Type II (failing to identify an impaired water) errors.

Nutrient concentrations less than or equal to the lower end (upper prediction interval) are unlikely to elicit the response threshold and therefore can be used as the basis for protective criteria, with a low probability of Type II statistical error but a high potential for Type I error. Conversely, a high likelihood of an undesirable response occurs when the nutrient concentration exceeds the upper end of the range (lower prediction interval). The probabilities of statistical errors at the upper end of the nutrient range are inverted compared with those at the lower end; that is, there is a low probability of Type I error and a higher probability for Type II error.

Because algal response is influenced by factors other than nutrients (grazing, macrophyte nutrient uptake, water retention time), the most scientifically defensible strategy for managing nutrients within the range of uncertainty is to verify a biological response before taking management action. If data demonstrate that a lake is biologically healthy and does not experience excess algal growth (e.g., < 20 µg chl. *a* /L in a colored lake or high specific conductance clear lake) despite having nutrient concentrations within the range of uncertainty, the existing nutrient concentrations appear to be acceptable. However, if the lake exhibits excess algal growth or biological impairment within this band of uncertainty, corrective action is warranted. Without chl. *a* data, FDEP should make decisions with much caution and assume an impaired condition if nutrients exceed the lower threshold. If chl. *a* data subsequently indicate that the designated use is indeed maintained at nutrient levels within the upper and lower prediction interval, those existing levels should be deemed acceptable.

Given this approach and using annual average chl. *a* values of 20 µg/L for colored lakes and higher-specific conductance clear lakes, and 6 µg/L for clear, low-specific conductance Florida lakes, respectively, criteria ranges associated with protection of designated uses can be defined on the basis of the 50% prediction intervals depicted in Figures 1-11 and 1-13. The resultant lower and upper thresholds of TN and TP for clear/low specific conductance lakes, clear/higher specific conductance lakes, and colored lakes are provided in Table 1-4.

Because color (PCU) is important in determining the applicable nutrient criteria, FDEP performed an analysis to establish the most appropriate averaging period for classifying a lake as clear or colored. For this analysis, FDEP obtained color data sets from several example lakes. Color data from multiple stations and years in a lake were then calculated as annual geometric means for each year. A rolling average was then calculated from the annual geometric means using varying periods to evaluate the period over which to average to minimize the variance in the resultant data set. Results indicate that a 5-year rolling average was generally sufficient to ensure minimization of the variance (for an example data set, see Figure 1-14).

1d. Duration and Frequency for Proposed Lakes Criteria

Numeric criteria include magnitude (quantity), duration, and frequency components. For the chl. *a*, TN, and TP criteria for lakes, the proposed criterion-magnitudes are expressed as annual geometric means, which includes duration (one-year averaging period). The criterion-frequency, or allowable excursion, is no more than once in a 3-year period. In addition, the long-term arithmetic average of annual geometric mean values may not exceed the criterion-magnitude values (concentration values).

Appropriate duration and frequency components of criteria should be based on how the data used to derive the criteria were analyzed and what the implications are for protecting designated uses given the effects of exposure at the specified criterion concentration for different periods and recurrence patterns. For lakes, the stressor-response relationship was based on annual geometric means for individual years at individual lakes. The appropriate period is therefore annual. The key question is whether this annual geometric mean needs to be met every year or if some allowance for a particular year to exceed the applicable criterion could still be considered protective.

Nutrient criteria, unlike criteria for toxic pollutants in most cases, are typically established within the range of natural variability. A temporary spike in nutrient levels does not necessarily harm the aquatic resource. In fact, natural systems have evolved to process variable inputs of nutrients, particularly where there is much natural variability in hydrologic conditions and precipitation patterns (e.g., wet years, dry years). Although biological response in the form of algal production, measured by chlorophyll *a*, can appear very quickly, longer term shifts in biological conditions, such as loss of underwater grasses, do not occur as the result of a single event or conditions in a single year. If grasses experience reduced light during a portion of a year, the effect on them is not expected to be as large or persistent as if reduced light occurs in multiple years. In other words, severe nutrient impacts often result from chronic exposure to elevated nutrients. In addition, extreme incidents of nutrient pollution are not likely to be isolated incidents. Severe exposure to high nutrients is most likely in the context of a water body that has chronic nutrient overenrichment.

Data that contributed to the analysis of TSI, as well as data generated from supporting paleolimnological studies and MEI modeling, typically represent periods greater than a single year. Conceptually, trophic state and the categories of oligotrophic, mesotrophic, and eutrophic, are attributes that are long term (longer than 1 year) and change with the long-term loading rates to a system. As an example, the eutrophication of Lake Washington and subsequent nutrient management and recovery of the lake were processes that took several decades (e.g., Edmondson 1994). Moreover, many of the models and analyses that form the basis of TSI results are designed to represent the "steady state," or long-term stable water quality conditions. Thus, the calculated criteria for lakes already have natural variability factored into them. They reflect central tendencies (with associated variability) of data, with decisions made about the distribution and variability surrounding those central tendency values.

However, researchers have suggested caution in applying steady-state assumptions to lakes with long residence times (Kenney 1990). In other words, the effects of spikes in annual loading could linger and disrupt the steady state in some lakes. As a result, U.S. EPA proposes two expressions of allowable frequency, both of which are to be met. First, U.S. EPA proposes a no more than one in 3 years excursion frequency for the annual geometric mean criteria for lakes. Second, U.S. EPA proposes that the long-term arithmetic average of annual geometric means not exceed the criterion-magnitude concentration.

The long-term average chl. *a* concentration is also associated with the probability of blooms with excessive chl. *a*. In a study of Vermont lakes (Walker 1984), a long-term average chl. *a* = 20 µg/L was associated with a 40% probability that instantaneous chl. *a* would be higher than 20 µg/L, and a 15% probability that instantaneous chl. *a* would be higher than 30 µg/L.

Those frequency and duration components take into account that hydrological variability will in turn produce variability in measured nutrient concentrations, and individual measurements may exceed the criteria. Furthermore, they balance the representation of underlying data and analyses on the basis of central tendency of many years of data (i.e., the long-term average component) with the need to exercise some caution to ensure that lakes have sufficient time to process individual years of elevated nutrient levels and avoid the possibility of cumulative and chronic effects (i.e., the no more than one in 3-year component).

1e. Application of Lake-specific Ambient Calculation Provision for Alternative TP and TN Criteria

U.S. EPA is proposing a framework that uses both the upper and lower bounds of the 50% prediction interval (Figures 1-11, 1-13) to allow the derivation of modified TP and TN lake-specific criteria to account for the natural variability in the relationship between chl. *a* response to TP and TN. The proposed rule would allow FDEP to calculate ambient modified criteria for TN and TP for a lake if chl. *a* is below the criterion magnitude in each of the 3 or more years of available ambient monitoring. Alternative criteria must be based on at least 3 years of ambient monitoring data with (a) at least four measurements per year and (b) at least one measurement between May and September and one measurement between October and April each year. If a calculated modified TN and TP criterion is below the lower bound (lower 50% prediction interval), the lower bound is the criteria. If a calculated modified TN and TP criterion is above the upper bound (upper 50% prediction interval), the upper bound is the criteria. Calculated modified TP and TN values may not exceed criteria applicable to streams to which a lake discharges.

The 50% prediction intervals and their derivation in the chl. *a* regression models are described above in Section 1c. The 50% prediction interval is the range within which one-half of chl. *a* observations are expected to fall for a given nutrient concentration (TN or TP), centered on the mean expectation at the regression line. In other words, the lower and upper bounds approximate the 25th and 75th percentiles of expected chl. *a* response for the given TN or TP, as predicted by the regression equations (Figures 1-11, 1-13).

One technical concern is the extent to which the variability in the data relating chl. *a* levels to TN and TP levels truly reflects differences among lakes, as opposed to temporal differences in the conditions in the same lake. In the data analysis leading up to this TSD, U.S. EPA analyzed individual lake data within the entire lake data set, to explore models of individual lake responses to nutrient enrichment (Appendix A-6). Models that incorporated individual lake response (single-lake models and Hierarchical Bayesian models that incorporate many lakes into the same model) verified that a large fraction of the total variability was due to "among lake" variability, and a small fraction was due to "within lake" variability (Appendix A-6, Figures A6-4).

Another technical concern is that a time lag might exist between the presence of high nutrients and the biological response. Lag time for changes of nutrient concentrations following change in loading is dependent on retention time, for example, twice the retention time has been observed (e.g., Coveney et al. 2005). Biological responses follow the nutrient lag time. Because of the potential lag time, U.S. EPA based the proposed range of TN and TP criteria on a 3-year averaging period so the time lag in response would not be expected to affect the ambient assessment.

A third technical concern is the presence of temporary or long-term, site-specific factors that could suppress biological response, such as the presence of grazing zooplankton, excess sedimentation that blocks light penetration, extensive canopy cover, or seasonal herbicide use that impedes proliferation of algae. If any of those suppressing factors are removed, nutrient levels could result in a spike in algal production above protective levels. Again, the 3-year data and averaging requirement will remove effects of temporary site-specific factors.

U.S. EPA proposes to require that the ambient calculation for modified TP and TN criteria be based on at least a 3-year record of observation and be based on representative sampling (i.e., four samples per year with at least one between May and September and one between October and April) during each year. Such requirements would minimize the influence of long-term, site-specific factors and ensure longer-term, stable conditions. U.S. EPA selected 3 years as a reasonable minimum length of time to appropriately account for anomalous conditions in any year that could lead to erroneous conclusions regarding the true relationship between nutrient levels in a lake and chl. *a* levels. U.S. EPA anticipates that the state would use all recent consecutive years of data on record (i.e., it would not be appropriate to select three random years within a complete record over the past seven years). U.S. EPA is requiring four measurements within a year to provide seasonal representation (i.e., May–September and October–April). Providing seasonal representation is important because nutrient levels can vary by season. In addition, this minimum sample size is conducive to deriving central tendency measurements, such as a geometric mean, with an acceptable degree of confidence. The chl. *a* criterion must be met in each of the three or more years of ambient monitoring that define the record of observation for the lake to be eligible for the ambient calculation modified provision for TN and TP.

1f. Alternatives Considered by U.S. EPA

i. Single Value Approach to Derive Lakes Criteria - Derive TN and TP Criteria Using Correlations Associated with the Regression Line or Upper Bound of the 50th Percentile Prediction Interval

As described in Section 1c, the proposed method uses the entire 50% prediction interval of the chlorophyll-nutrient regressions to derive the criteria (Figures 1-11, 1-13). The point at which the *upper*-prediction bound intersects with the chlorophyll criterion (6 or 20 µg/L chl. *a*) corresponds to *lower*-nutrient concentrations (TP and TN; Figures 1-11, 1-13). In terms of risk, the upper-prediction bound corresponds to an estimated risk that 25% of lakes with the given nutrient concentration will exceed the chl. *a* criterion, and the lower bound corresponds to a risk that 75% of lakes with that concentration of nutrient will exceed the chl. *a* criterion. The default criterion is the upper bound (25% risk), but if TN or TP exceed that limit yet chl. *a* remains below the criterion, TN and TP criteria might be higher, up to that corresponding to 75% risk.

An alternative is to use a single criterion of the upper bound or the regression line, with no option of alternative TN or TP criteria. These alternatives are more conservative and fix the TN and TP criteria to an estimated 25% risk of chl. *a* exceeding the chl. *a* criterion if the upper bound is used, or an estimated 50% risk of chl. *a* exceeding the chl. *a* criterion if the regression line is used. Given the variability among lakes (see Appendix A-6), the upper bound criterion could result in TN and TP criteria more stringent than absolutely necessary for some lakes. Alternatively, the regression line criterion could result in TN and TP criteria that represent a balance of risk.

U.S. EPA's rationale for proposing a framework that uses both the upper and lower bounds of the 50th percentile prediction interval to allow the derivation of modified TN and TP lake-specific criteria rather than either of these single values was to account for the natural variability in the relationship between chl. *a* and TN and TP that can exist in lakes.

ii. Additional Modification to Proposed Lakes Classification

U.S. EPA examined alkalinity in clear Florida lakes to identify alternatives to the classification described in Section 1f.i. Single measurements of nutrients and chl. *a* re highly variable and can lead to results that cannot be interpreted, so annual geometric mean concentrations were used as the data units for the analysis. The data requirements are: a minimum of four samples per year, and at least one sample in each of the cool season (October to April) and the warm season (May to September). That resulted in 492 annual geometric means from 180 lakes. Individual lakes had 1–12 annual means. Alkalinity has been measured less frequently than nutrients or chlorophyll. Approximately half of the lakes had associated alkalinity observations, resulting in a total of 200 annual means from 89 lakes for this analysis.

The distribution of log alkalinity in Florida clear lakes (Figure 1-15) shows a peak of very low alkalinities less than 1 mg/L (negative log values, Figure 1-15), a range of intermediate, low alkalinities, and a second peak at 50–90 mg/L. Lakes with alkalinity less than 1 mg/L are essentially unbuffered, highly acidic lakes. In this distribution, the alkaline lakes appear as a group with alkalinities above 20 mg/L.

TP, TN, chlorophyll, and specific conductance are all correlated with alkalinity in the described data set (Figure 1-16). Change-point analysis showed change points at alkalinities of 13–24 mg/L. Associations of chlorophyll and TN with alkalinity were the strongest, such that acidic lakes were associated with low values of TN and chlorophyll. As an alternative classification, the alkalinity breakpoint could be set at 20 mg/L to reflect similar breakpoints in chlorophyll and nutrient concentrations.

As described in Section 1a, the response of chlorophyll to nutrient concentrations was not different between acid and alkaline lakes (Figures 1-5, 1-17). The acidic and alkaline lakes appeared to lie on the same regression relationship, although alkaline lakes had higher mean nutrient and chlorophyll concentrations. Acidic lakes showed considerably more variability around the regression line, especially in the chlorophyll-TN regression (Figure 1-4). Figure 1-5 shows the alkalinity classification breakpoint at 50 mg/L $CaCO_3$. If the class breakpoint is at 20 mg/L, as suggested by the change point analysis, the separation of the alkalinity classes is more distinct (Figure 1-17).

Alkalinity and Specific Conductance

U.S. EPA examined whether specific conductance can be used as a surrogate for alkalinity in classifying Florida lakes, because alkalinity is not universally available in historical data. Data used were the same data from FDEP described at the beginning of this section.

In Florida's lake database, only a limited number of lakes (386) were measured for alkalinity while the majority of the lakes were measured for specific conductance. Because of its strong correlation with alkalinity and large sample size, specific conductance was considered as a surrogate for alkalinity. Figure 1-18 (a) indicates a strong correlation between specific conductance and total alkalinity in clear lakes (Spearman $R = 0.871$). A change point analysis indicated that a change point occurs when specific conductance is around 100 µS/cm. Nonlinear median quantile regression was also used to examine the relationship between specific conductance and alkalinity (Figure 1-18 (b)). FL has proposed to use 50 mg/L as a threshold for determining alkaline and acidic waters. In the analysis, the intercept of 50 mg/L alkalinity with the quantile regression line was around a specific conductance value of 200 µS/cm (Figure 1-18 (b)). The alternative alkalinity classification breakpoint at 20 mg/L total alkalinity yields a specific conductance breakpoint at approximately 100 µS/cm, equivalent to the change point analysis result.

iii. Modification to Include Upper Percentile Criteria with an Associated Ten Percent Exceedance Frequency

Variance Components

In developing nutrient criteria, including frequency and duration components, U.S. EPA also considered upper percentile criteria with low exceedance frequencies in addition to the annual average criteria concentrations. The upper percentile values analyzed here are in addition to the annual averages, as an alternative.

The objective of this analysis was to assign a criterion for single nutrient observations that would be protective of an annual mean nutrient concentration while taking into account natural variability. To predict nutrient concentrations in any particular lake, two types of variation must be considered: within lake variation and among lake variation. Because some lakes have been sampled hundreds of times, and others might have been sampled very few times, sampling errors within lakes could be variable.

U.S. EPA calculated within-lake variance for each lake and then plotted the cumulative distribution function (Figure 1-19). The mean of the observed standard deviation distribution was used as the estimated standard deviation for the population. The mean standard deviations of TP for acidic and alkaline lakes were 0.514 and 0.451, respectively (Table 1-5).

A second approach used was hierarchical modeling to determine population variation. Hierarchical modeling has been applied frequently to balance within and among group variations. The basic idea of hierarchical modeling (also known as multilevel modeling, empirical Bayes, random coefficient modeling, or growth curve modeling) is to think of the lowest-level units (smallest and most numerous) as organized into a hierarchy of successively higher-level units. For example, TP concentrations are in lakes, lakes are in regions, and regions are in lake classes. Outcomes for individual lakes can be described as a sum of effects for the individual observations, for the observations of the lake, and for the whole region. Each of these effects can often be regarded as one of an exchangeable collection of effects (e.g., all region-level effects) drawn from a distribution described by a variance component. Once a model of this type is specified, inferences can be drawn from available data for the population means at any level (lake, region, class, etc.) (Price et al. 1996).

These estimators, which can be regarded from a Bayesian perspective as posterior means or from a frequentist perspective as "Best Linear Unbiased Predictors" often have better properties than simple sample-based estimators using only data from the unit in question. This makes them useful in the problem of "small-area estimation," i.e., making estimates for units or domains for which there is a very limited amount of information (Osborne 2000).

The R package lme4 was used for the hierarchical mixed modeling. The computed means and standard deviations (SDs) are also summarized in Table 1-5.

Distribution

Because the population mean and SD are known (the proposed mean TP criterion for acidic lakes is 10 mg/L, and for alkaline lakes is 30 mg/L), the probability of a random sample occurring within a normal distribution is known (Figure 1-20), and thus values were calculated for exceeding the selected probability. In other words, when a single TP value falls at the right side of the vertical line, there is only less than 5% probability that the mean TP concentration at that lake would have a mean of 10 mg/L (acidic) or 30 mg/L (alkaline) or less. U.S. EPA used both the mean observed SD and modeled SD values as the population SD to generate single observation criteria (Table 1-6). The modeled SD are similar to the average observed SD-based single criteria most of the time.

Table 1-1. Warm-water TSI Categories (after Salas and Martino 1991)

TSI	Category	TP (µg/L)	Chl. *A* (µg/L)
40	Oligotrophic	21.3	5
50	Mesotrophic	39.6	10
70	Eutrophic	118.7	40

Table 1-2. Relationship between Chl. *a*, TP, and TN, as Described by Florida's TSI

TSI	Chl. *a* (µg/L)	TP (mg/L)	TN (mg/L)
0	0.3	0.003	0.06
10	0.6	0.005	0.10
20	1.3	0.009	0.16
30	2.5	0.01	0.27
40	5.0	0.02	0.45
50	10.0	0.04	0.70
60	20	0.07	1.2
70	40	0.12	2.0
80	80	0.20	3.4
90	160	0.34	5.6
100	320	0.58	9.3

Table 1-3. Summary of the Linear Multiple Regression between Ln Transformed Chl. *a* and Ln Transformed TP and TN in Colored Florida Lakes

Effect	Coefficient	Std Error	Std Coef.	Tolerance	T	P (2 tail)
Constant	2.703	0.272	0		9.922	0.0000
LTP	0.347	0.085	0.213	0.457	4.091	0.0000
LTN	1.546	0.149	0.542	0.457	10.395	0.0000

Source	Sum-of-squares	DF	Mean-square	F-ratio	P
Regression	449.764	2	224.882	205.223	0.0000
Residual	432.839	395	1.096		

Note: The regression multiple R^2 and adjusted R^2 were 0.510 and 0.507, respectively.

Table 1-4. TP and TN Criteria Ranges for Clear (< 40 PCU) and Colored Florida Lakes (> 40 PCU)

A	B	C	D	E	F
Long-term Average Lake Color and Alkalinity	Chl. *a* [f] (µg/L) [a]	Baseline criteria [b]		Alternative criteria (within these bounds) [c]	
		TP (mg/L) [a]	TN (mg/L) [a]	TP (mg/L) [a]	TN (mg/L) [a]
Colored Lakes > 40 PCU	20	0.050	1.23	0.050–0.157	1.23–2.25
Clear Lakes, Alkaline ≤ 40 PCU [d] and > 50 mg/L $CaCO_3$ [e]	20	0.030	1.00	0.030–0.087	1.00–1.81
Clear Lakes, Acidic ≤ 40 PCU [d] and ≤ 50 mg/L $CaCO_3$ [e]	6	0.010	0.500	0.010–0.030	0.500–0.900

Note: The lower and upper thresholds were based on the intersection of chl. *a* response concentrations with the 50% predictions intervals shown in Figures 1-11 and 1-13.

[a] Criteria values are based on annual geometric mean not to be exceeded more than once in a 3- year period. (Duration = annual; Frequency = not to be exceeded more than once in a 3-year period).

[b] Baseline criteria apply unless other data are readily available to calculate and apply as alternative criteria as described below in footnote c and Florida decides to implement and publicly certify them as an alternative WQS on the record as documented in an easily accessible and publicly available location, such as an official state Web site.

[c] If chl. *a* is below the criterion in column B and there are representative data to calculate ambient TP and TN criteria, alternative criteria may be calculated within these bounds from ambient measurements to determine lake-specific criteria pursuant to CWA section 303(c). The calculated ambient condition TN and TP values must be based on at least three years of ambient monitoring data with (a) at least four measurements per year and (b) at least one measurement between May and September and one measurement between October and April each year. These same data requirements apply to chl. *a* when determining whether the chl. *a* criterion is met for purposes of conducting the ambient calculation. If the calculated TN and TP value is below the lower bound, the lower bound is the criterion. If the calculated TN and TP value is above the upper bound, the upper bound is the criterion. Calculated TP and TN values may not exceed criteria applicable to streams to which a lake discharges. If chl. *a* is below the criterion in column B and representative data to calculate ambient TP and TN criteria are not available, then the baseline TN and TP criteria apply.

[d] Platinum Cobalt Units assessed as true color free from turbidity. Long-term average color based on a rolling average of up to 7 years using all available lake color data.

[e] If alkalinity data are unavailable, a specific conductance of 250 micromhos/cm may be substituted.

[f] Chl. *a* is defined as corrected chlorophyll, or the concentration of chl. *a* remaining after the chlorophyll degradation product, phaeophytin *a*, has been subtracted from the uncorrected chl. *a* measurement.

Table 1-5. Summary Statistics (Mean and SD) of Nutrient Variables for Acidic, Alkaline Lakes, and Colored Lakes, Meeting Annual Criteria

	Acidic Lakes			**Alkaline Lakes**			**Colored Lakes**		
	ln TP	**ln TN**	**ln Chl. *a***	**ln TP**	**ln TN**	**ln Chl. *a***	**ln TP**	**ln TN**	**ln Chl. *a***
Criterion	–4.605	–0.693	1.792	–3.507	0.000	2.996	–2.996	0.182	2.996
Obs Mean	–5.045	–1.215	0.123	–4.212	–0.414	0.872	–3.779	–0.218	1.200
Obs Mean SD	0.518	0.204	0.730	0.451	0.451	0.675	0.416	0.185	0.889
Mod Mean	–5.031	–1.221	0.213	–4.121	–0.387	0.965	–3.790	–0.211	1.263
Mod SD	0.586	0.293	0.884	0.544	0.201	1.093	0.513	0.262	1.095

Table 1-6. Single Observation Criteria for Nutrient Variables Derived Using Both Mean Observed SD and Modeled SD, for Lakes Meeting Respective Criteria

	Acidic Lakes			Alkaline Lakes			Colored Lakes		
	TP (mg/L)	TN (mg/L)	Chl. *A* (µg/L)	TP (mg/L)	TN (mg/L)	Chl. *A* (µg/L)	TP (mg/L)	TN (mg/L)	Chl. *A* (µg/L)
Obs 90th	0.019	0.649	15	0.053	1.782	48	0.085	1.521	63
Mod 90th	0.021	0.728	19	0.060	1.294	81	0.096	1.678	81
Obs 95th	0.023	0.699	20	0.063	2	61	0.099	1.626	86
Mod 95th	0.026	0.810	26	0.073	1.392	121	0.116	1.846	121
Obs 97.5th	0.027	0.74	25	0.07	2.42	75	0.11	1.72	114
Mod 97.5th	0.032	0.888	34	0.087	1.483	170	0.137	2.005	171
Obs 99th	0.033	0.804	33	0.086	2.855	96	0.132	1.845	158
Mod 99th	0.039	0.989	47	0.106	1.596	254	0.165	2.207	256

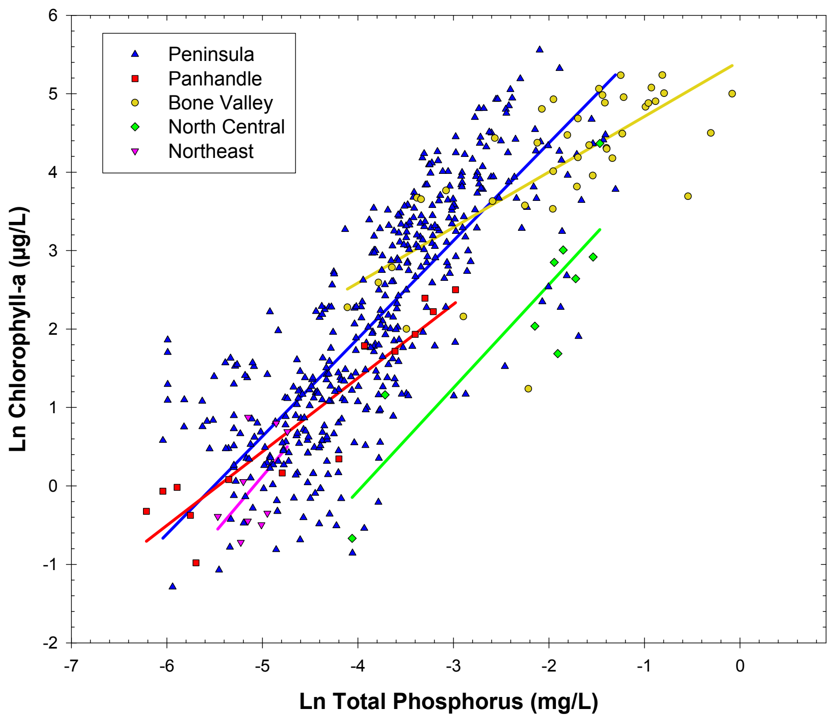


Figure 1-1. Relationship between Ln transformed chl. *a* and TP in clear lakes by nutrient region. Note, over the entire range of data, that the lakes exhibit a similar chlorophyll response to TP independent of region.

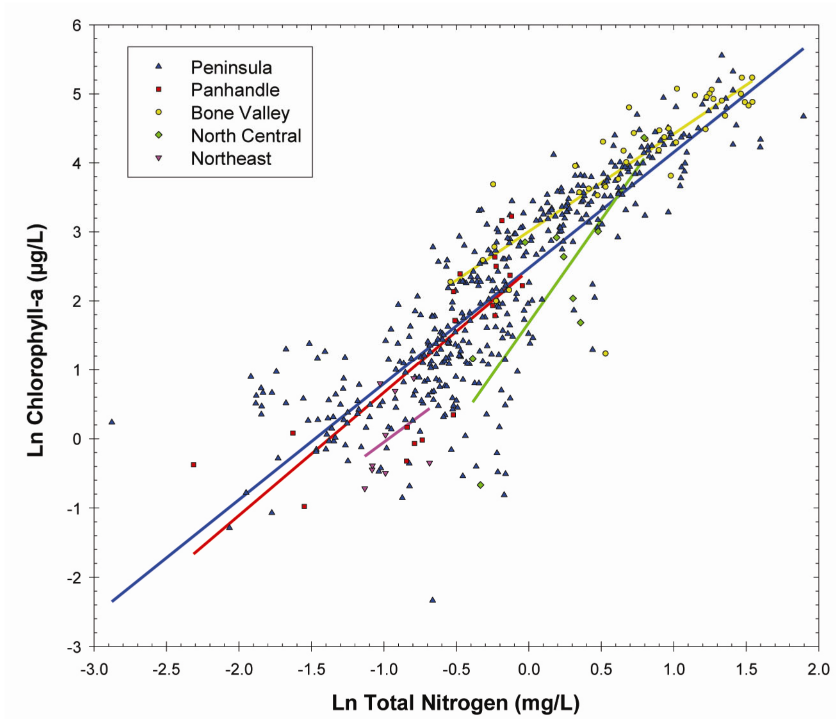


Figure 1-2. Relationship between Ln transformed chl. *a* and TN in clear lakes by nutrient region. Note, over the entire range of data, that the lakes exhibit a similar chlorophyll response to TN independent of region.

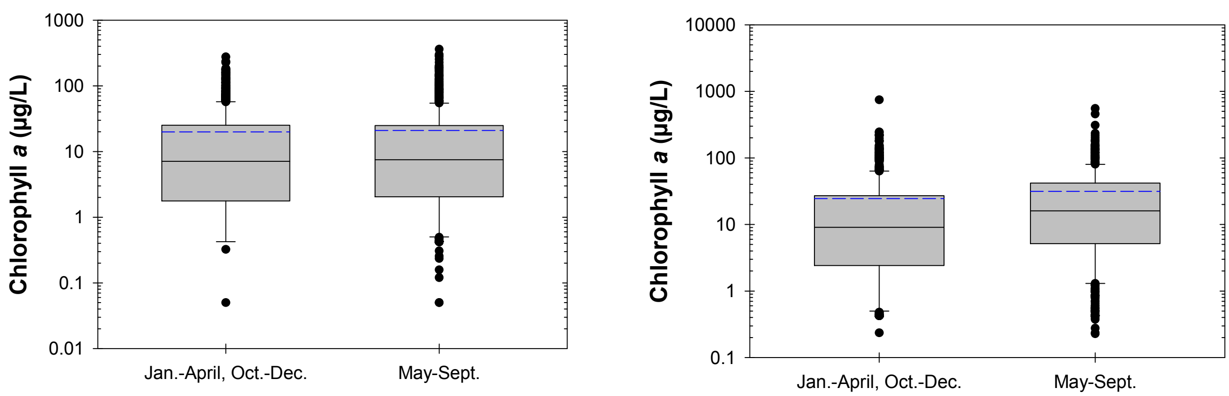


Figure 1-3. Box plots of chl. *a* by season in clear (left panel) and colored (right panel) lakes.

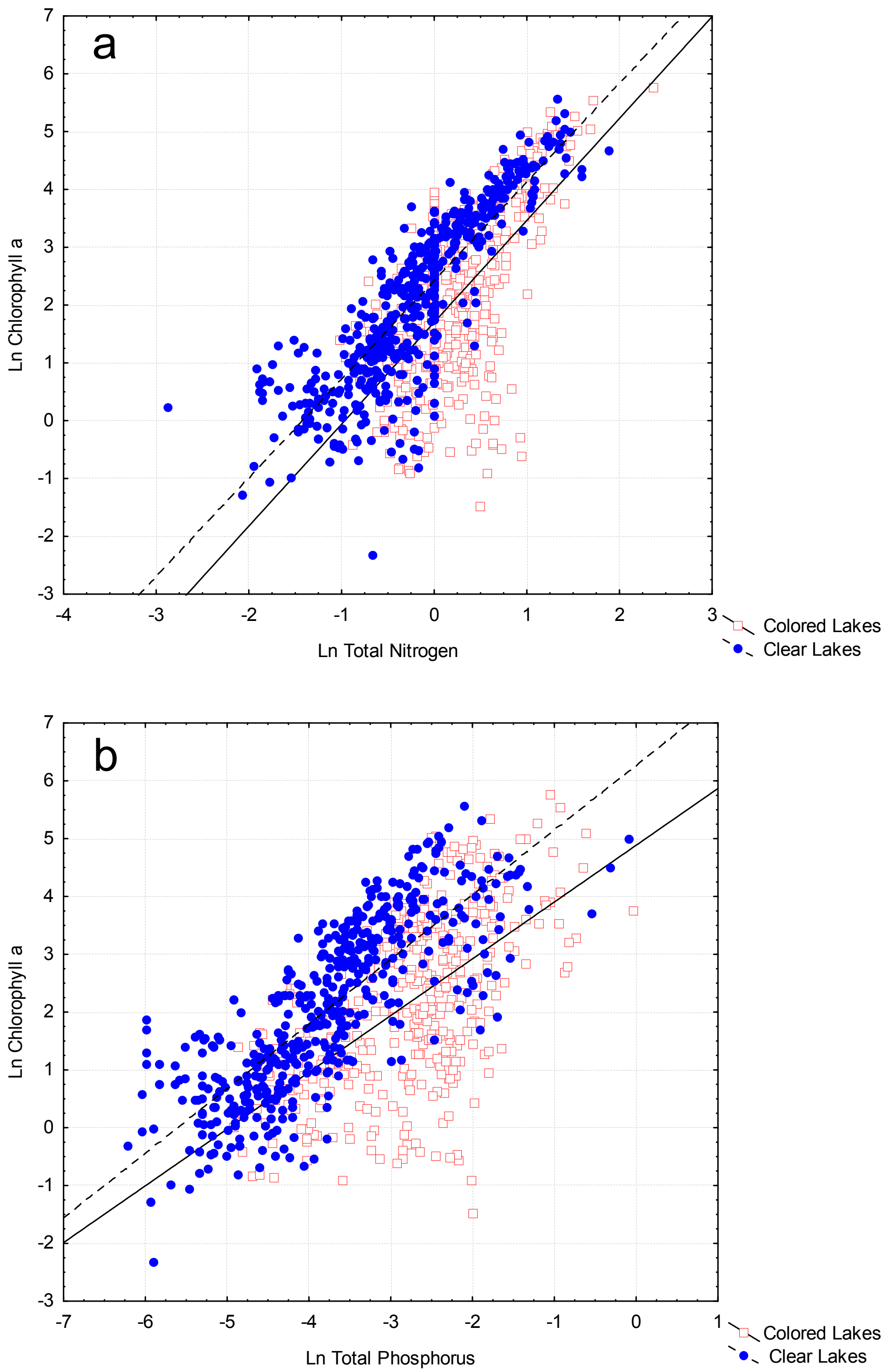


Figure 1-4. Regression analyses of annual geometric mean chl. *a* concentration and nutrients in colored and clear lakes, showing grouping of the two lake classes and the different regression lines.

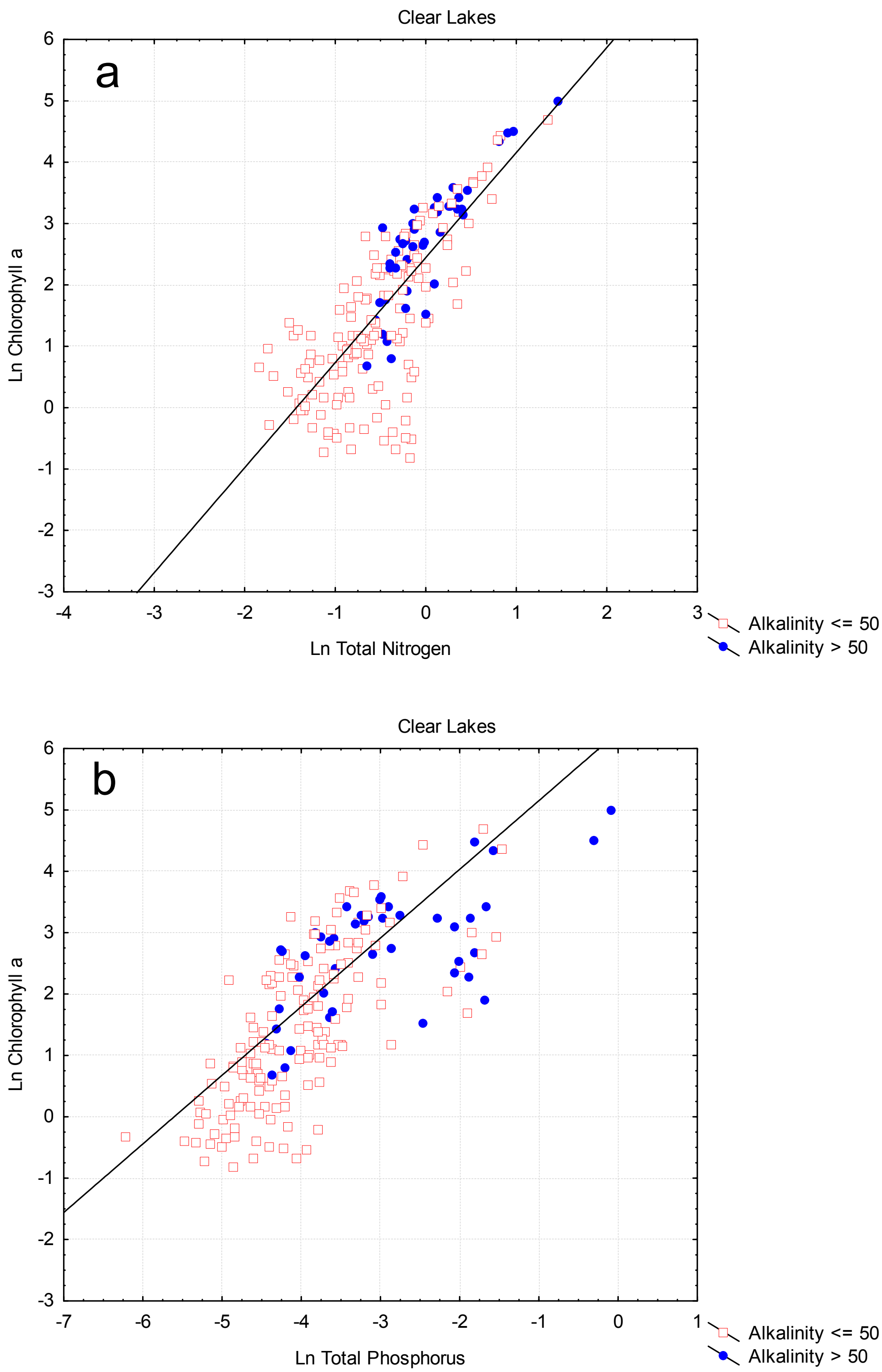


Figure 1-5. Scatter plot of annual geometric mean chl. *a* concentration and nutrients of clear lakes, showing acidic (< 50 mg/L $CaCO_3$) and alkaline lakes. Acidic and alkaline groups were combined in a single regression.

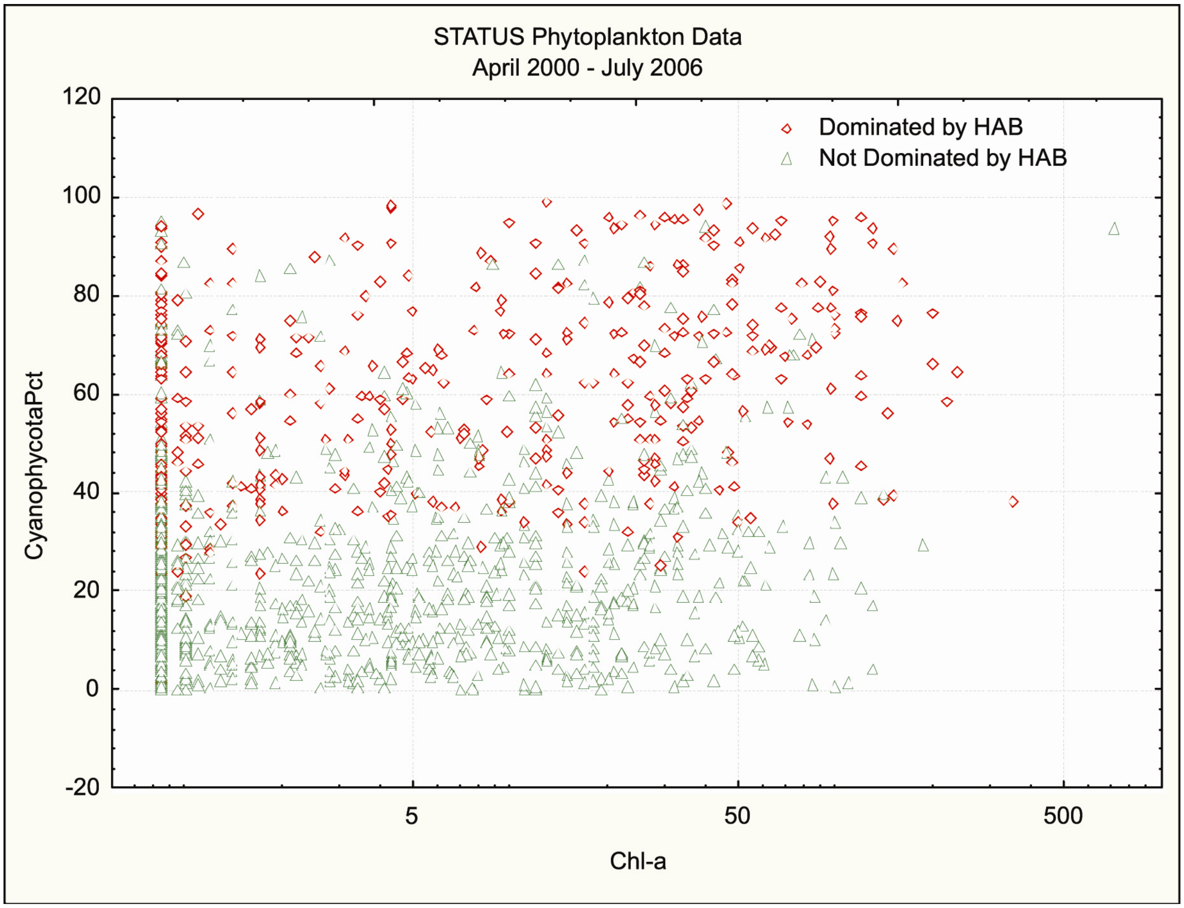


Figure 1-6. The relationship between chl. *a* concentrations (note the log scale) and the percent cyanobacteria in 1,364 lake samples collected for Florida's statewide probabilistic monitoring program.

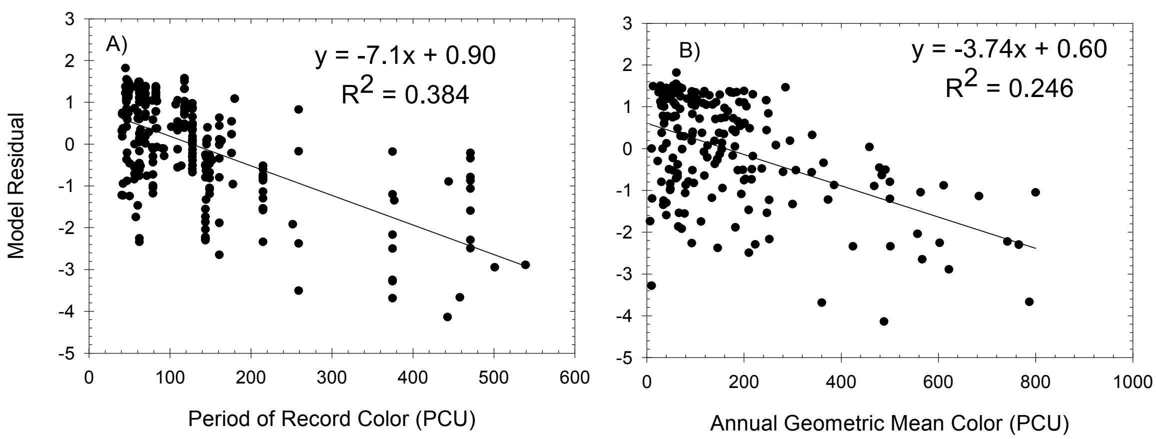
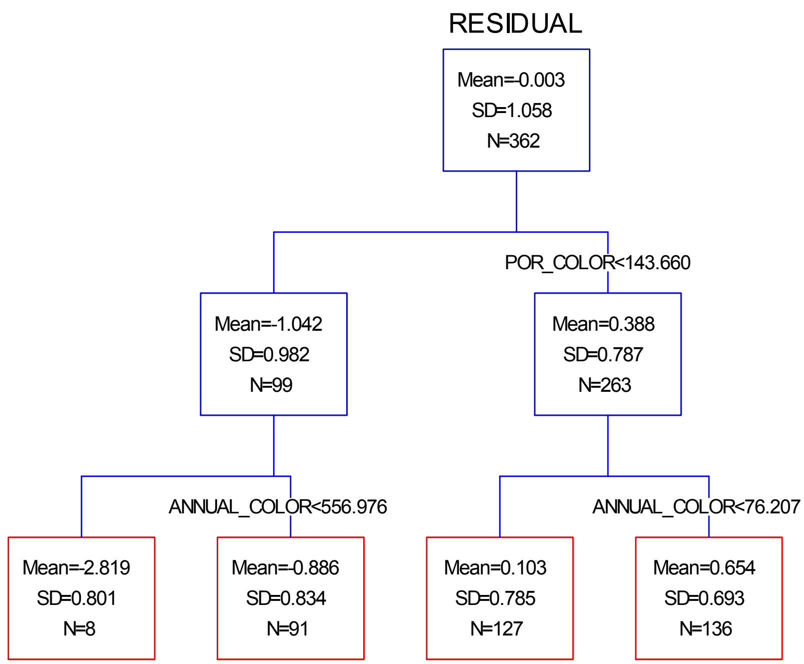


Figure 1-7. Relationship between the residual error in the TP and TN chlorophyll model and (A) period of record geometric mean lake color and (B) annual geometric mean lake color. Note that both relationships exhibit a significant negative slope.



Split	Variable	PRE*	Improvement
1	POR_COLOR	0.333	0.333
2	Annual_COLOR	0.371	0.039
3	Annual_COLOR	0.409	0.037

Figure 1-8. CART analysis, using a least-squares fitting method, of the residual error from the TP and TN model for chl. *a* response in colored Florida lakes. The analysis demonstrates that colored lakes can be split into two large groups, using the first CART split, where the chl. *a* response to TP and TN differs because of the confounding effect of color. *Proportion reduction in error.

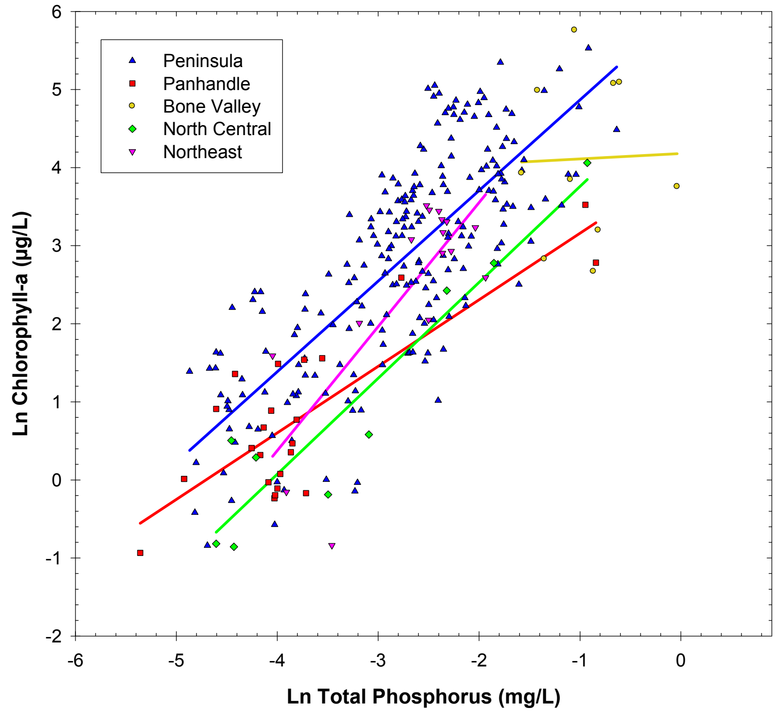


Figure 1-9. Relationship between Ln transformed chl. *a* and TP in moderately colored lakes by nutrient region. Note, over the entire range of data, that the lakes exhibit a similar chlorophyll response to TP independent of region.

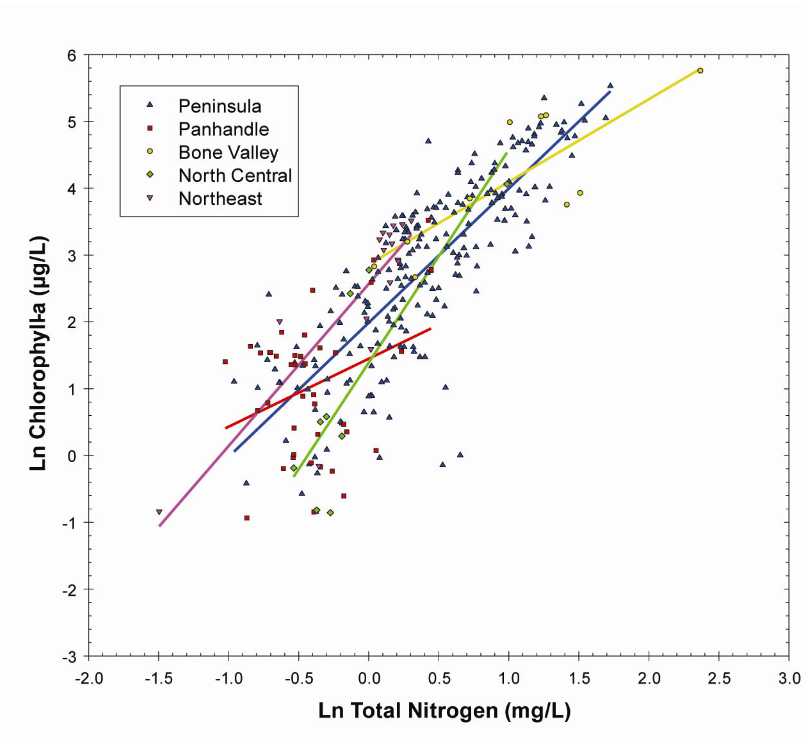


Figure 1-10. Relationship between Ln transformed chl. *a* and TN in moderately colored lakes by nutrient region. Note, over the entire range of data, that the lakes exhibit a similar chlorophyll response to TN independent of region.

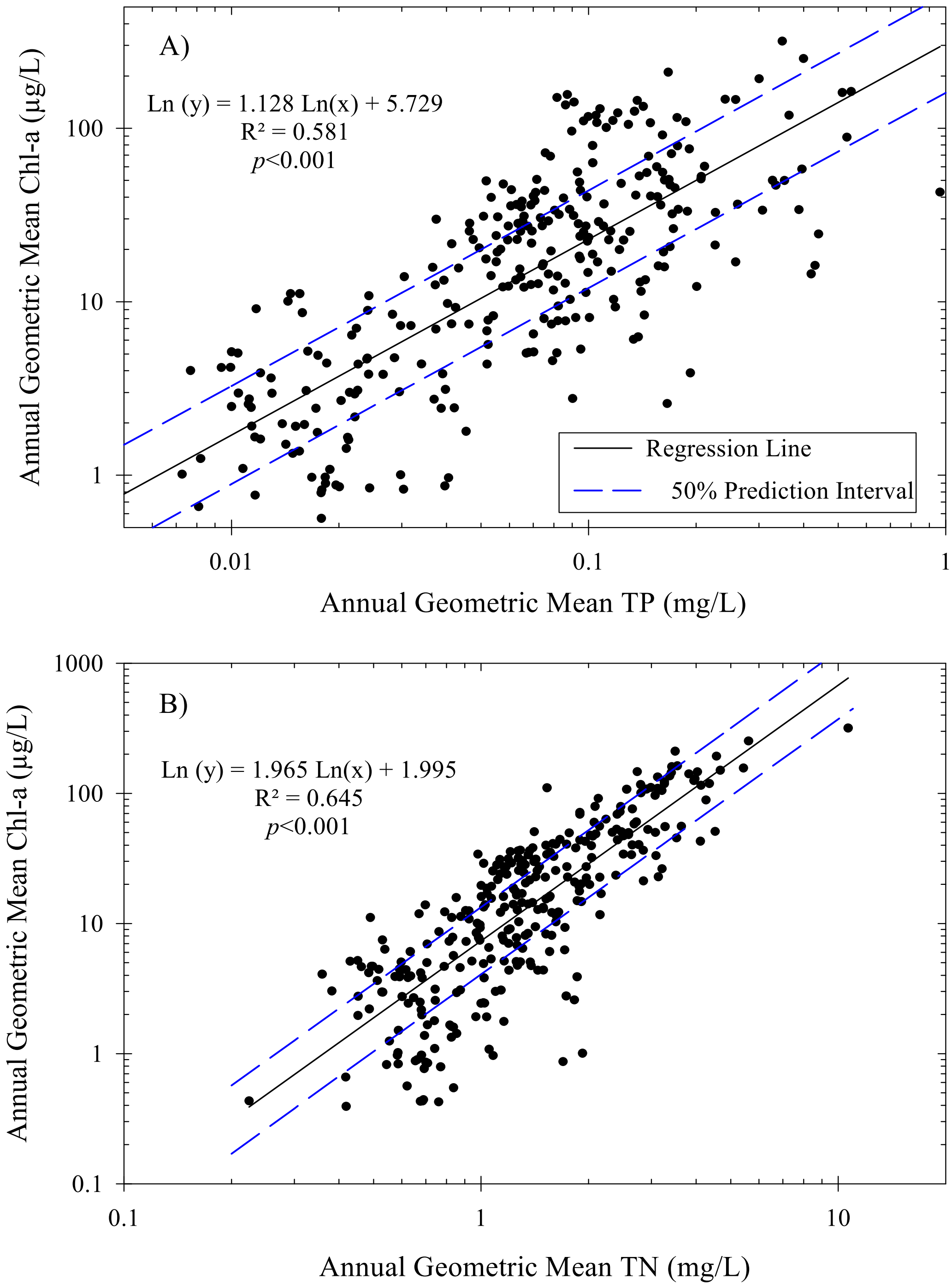


Figure 1-11. Regression analyses annual geometric mean chl. *a* concentrations and annual geometric mean (A) TP and (B) TN concentrations in moderately colored (> 40–140 PCU) Florida lakes. Note that the x axis and y axis are both expressed on a log-scale.

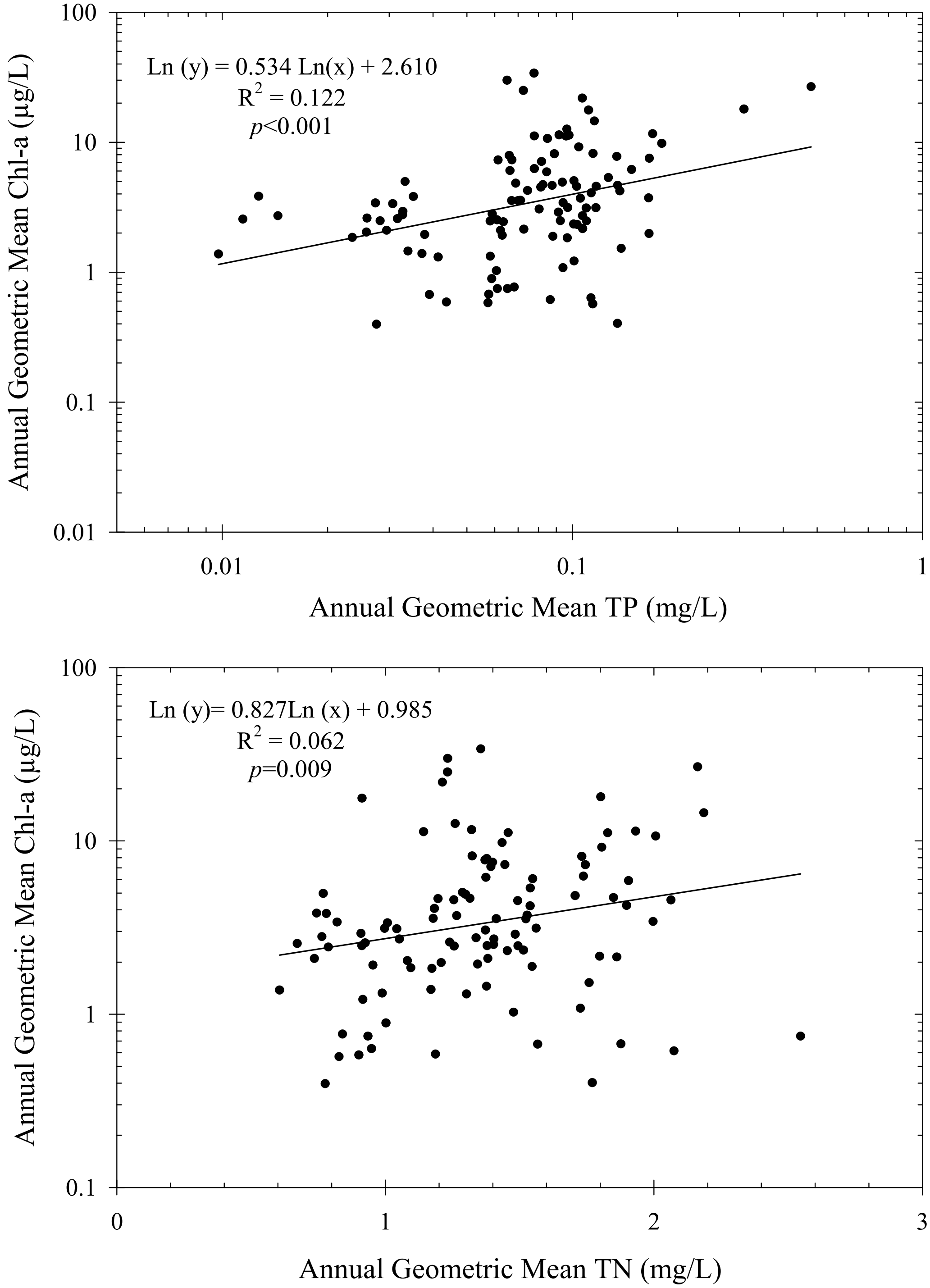


Figure 1-12. Regression analyses annual geometric mean chl. *a* concentrations and annual geometric mean (A) TP and (B) TN concentrations in highly colored (> 140 PCU) Florida lakes. Note that the x axis and y axis are both expressed on a log-scale.

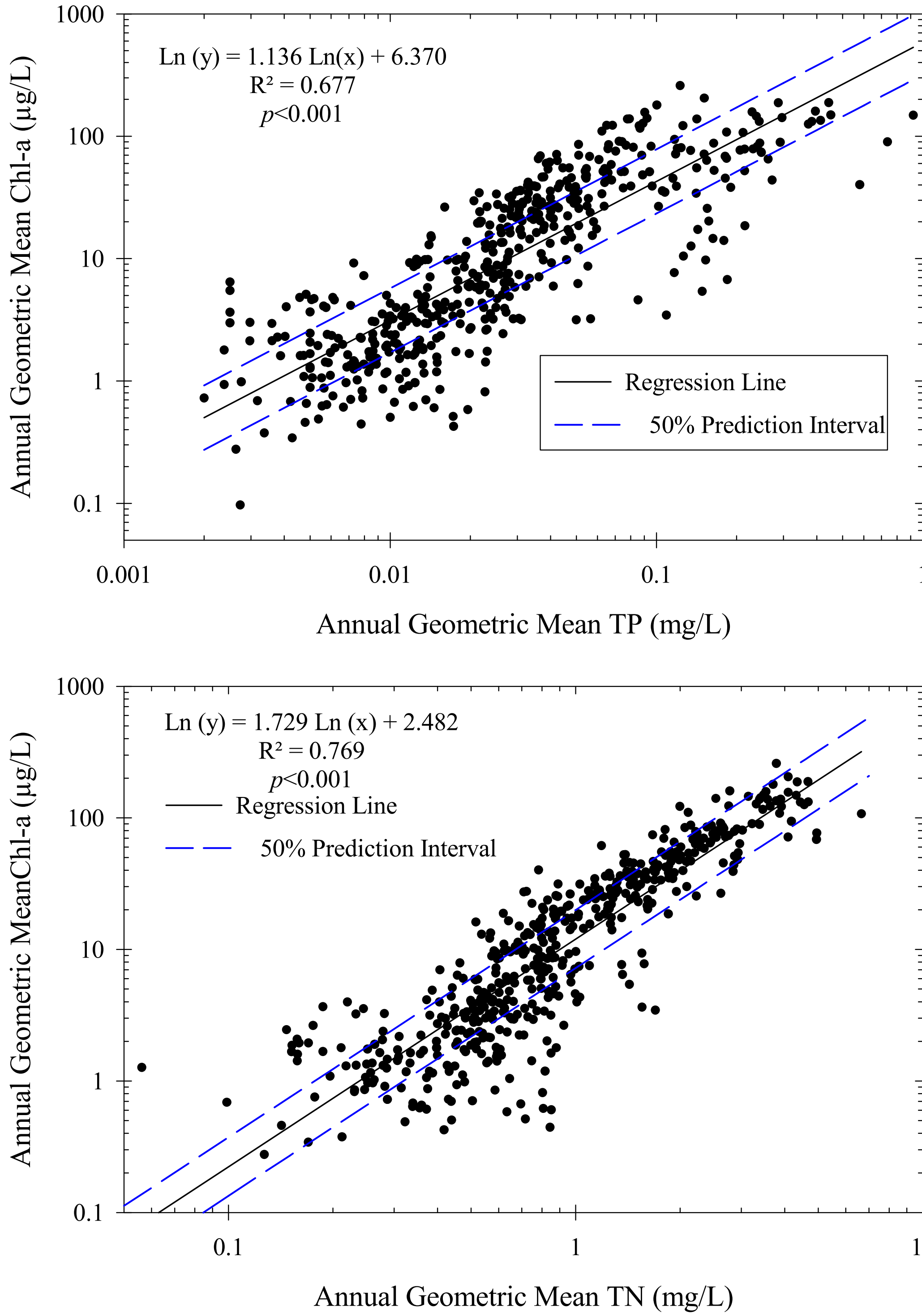


Figure 1-13. Regression analyses annual geometric mean chl. *a* concentrations and annual geometric mean (A) TP and (B) TN concentrations in clear Florida lakes. Note that x axis and y axis are both expressed on a log-scale.

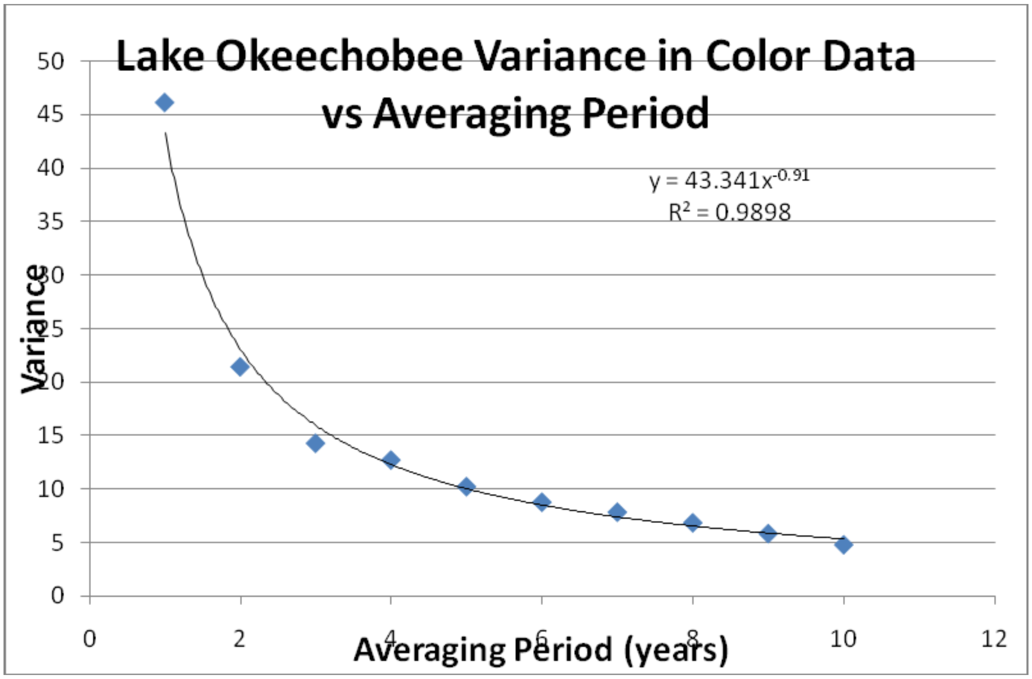


Figure 1-14. Variance in Lake Okeechobee color data from eight pelagic stations, averaged over varying time periods.

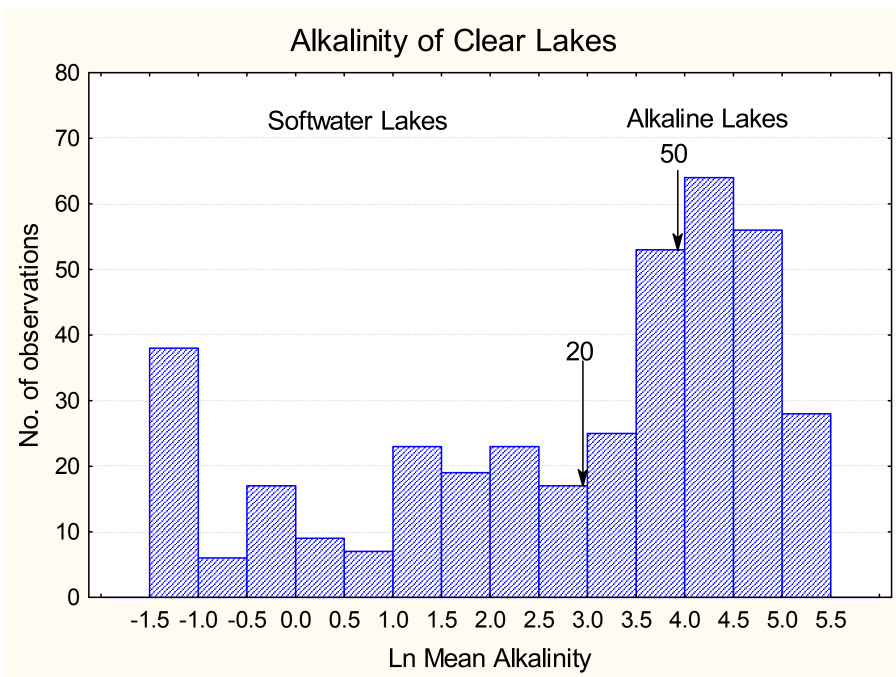


Figure 1-15. Distribution of average log alkalinity in clear Florida lakes (log scale). Alkalinities of 20 and 50 mg/L shown by arrows.

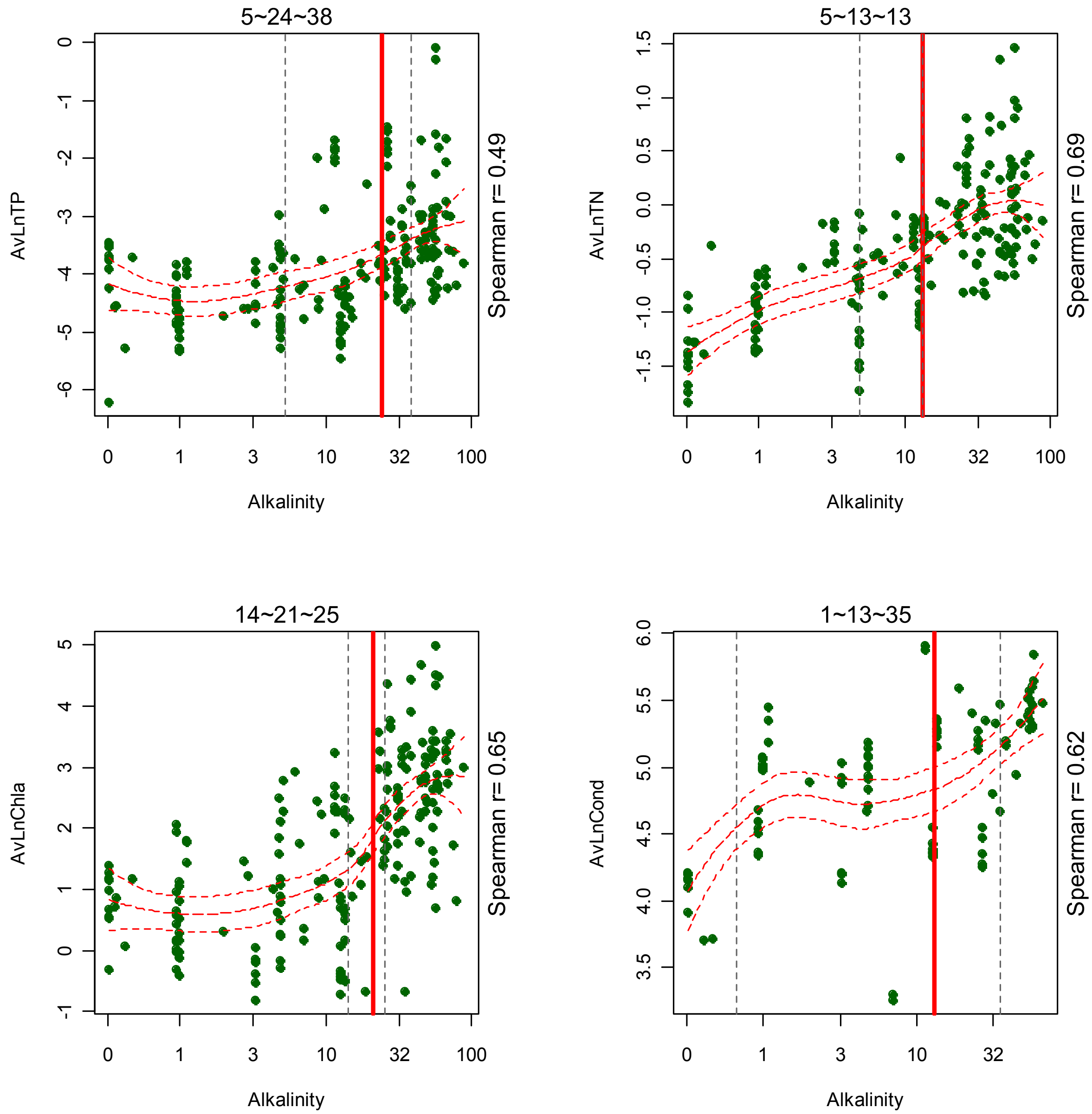


Figure 1-16. Scatterplots of mean TP, TN, Chl. *a*, and specific conductance on lake alkalinity. Vertical solid lines are the estimated change points, and vertical dashed lines denote the 95% confidence intervals for the change points. Wavy dashed lines are LOESS nonlinear regressions and confidence intervals.

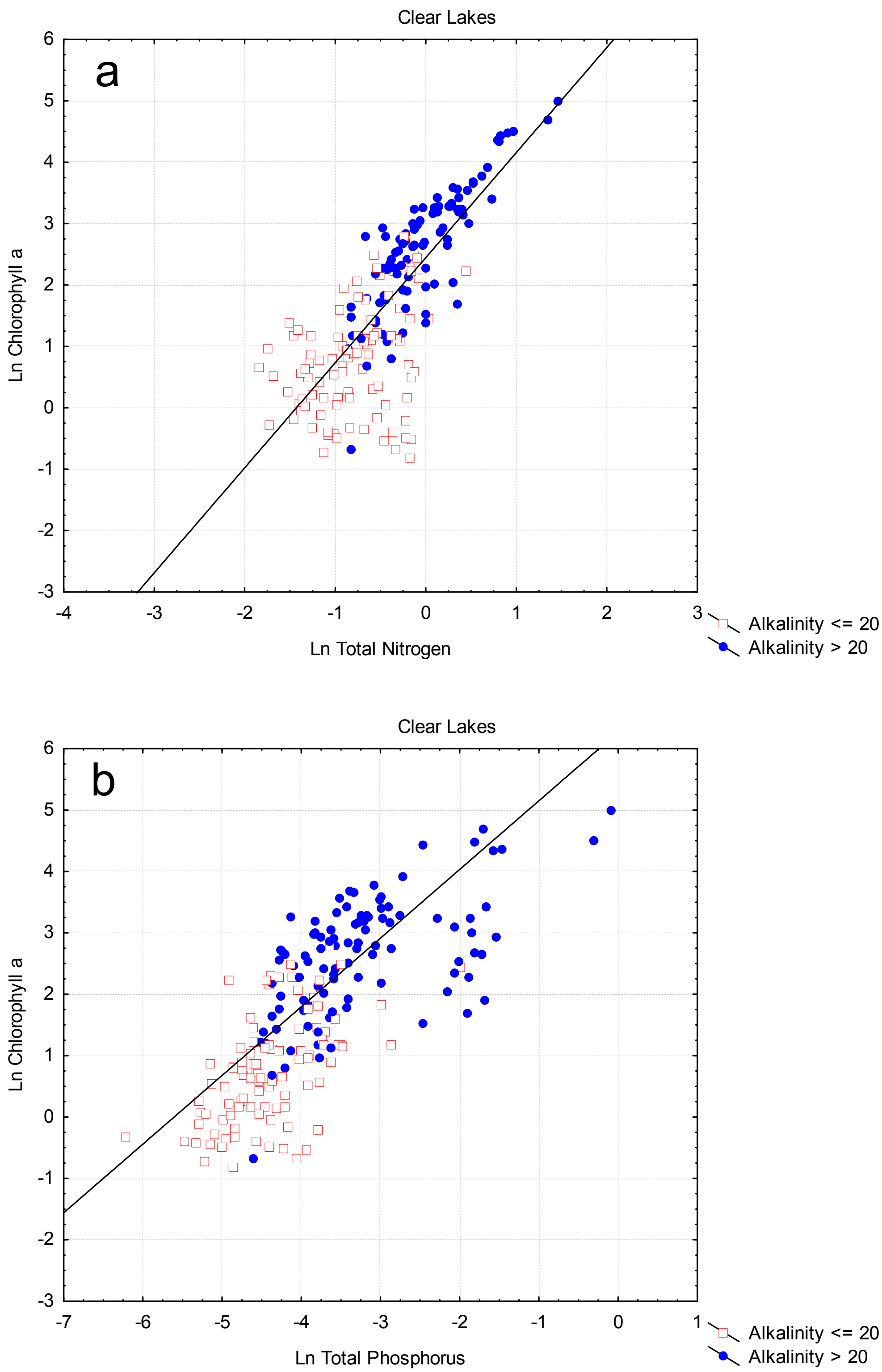


Figure 1-17. Scatter plot of annual geometric mean chl. *a* concentration and nutrients of clear lakes, showing separation of acidic and alkaline lakes with classification breakpoint at 20 mg/L $CaCO_3$. Compare to Figure 1-5. Acidic and alkaline groups were combined in a single regression. a: TN; b: TP.

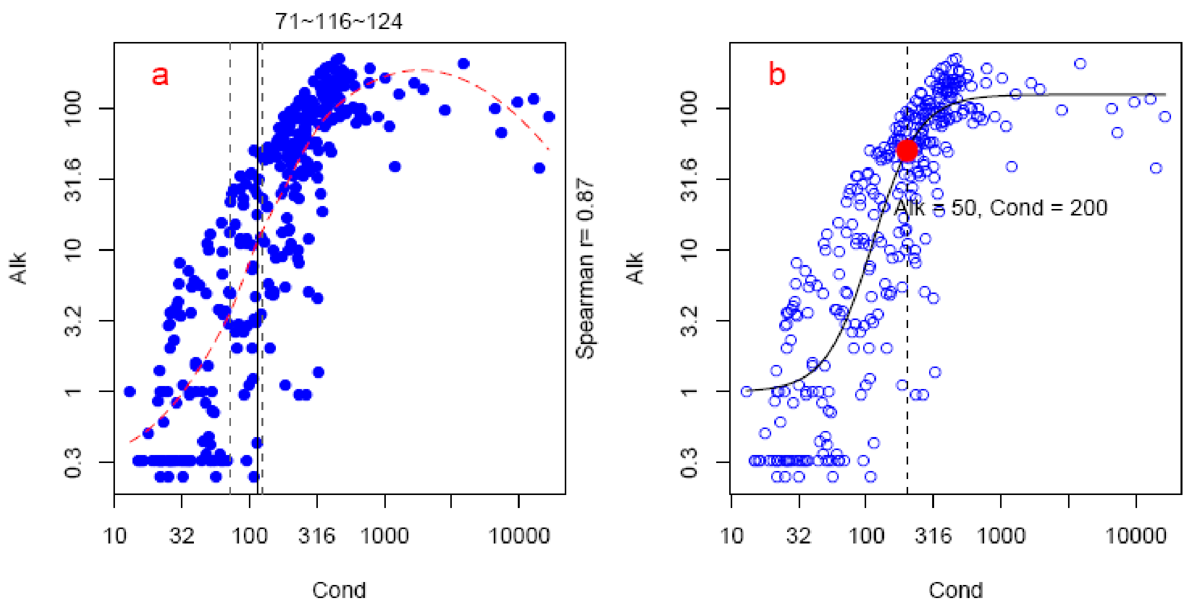


Figure 1-18. Relationship between specific conductance and alkalinity in clear lakes. (a) The curve shows loess fitted line, and the vertical lines show change point and its 90% confidence limits; (b) The curve shows median quantile fit, and the red dot is the intercept of the quantile regression line with specific conductance = 200 µS/cm.

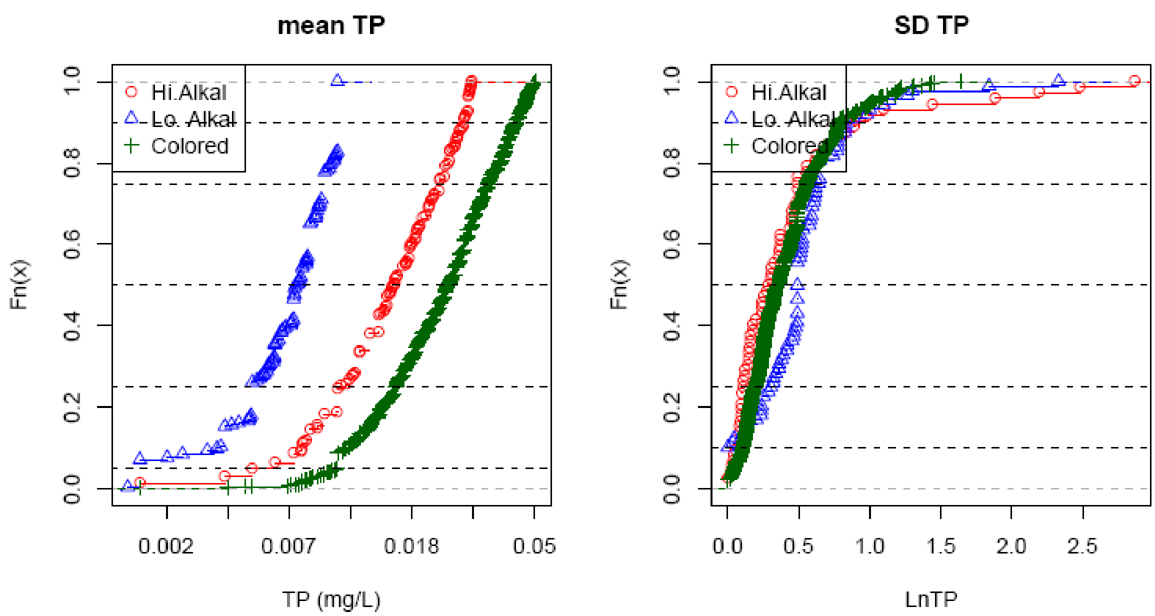


Figure 1-19. Cumulative frequency distribution of mean TP concentrations in acidic and alkaline lakes that meet the annual average criteria. A specific conductance > or < 250 µS/cm is applied as a threshold for classifying high- or acidic lakes.

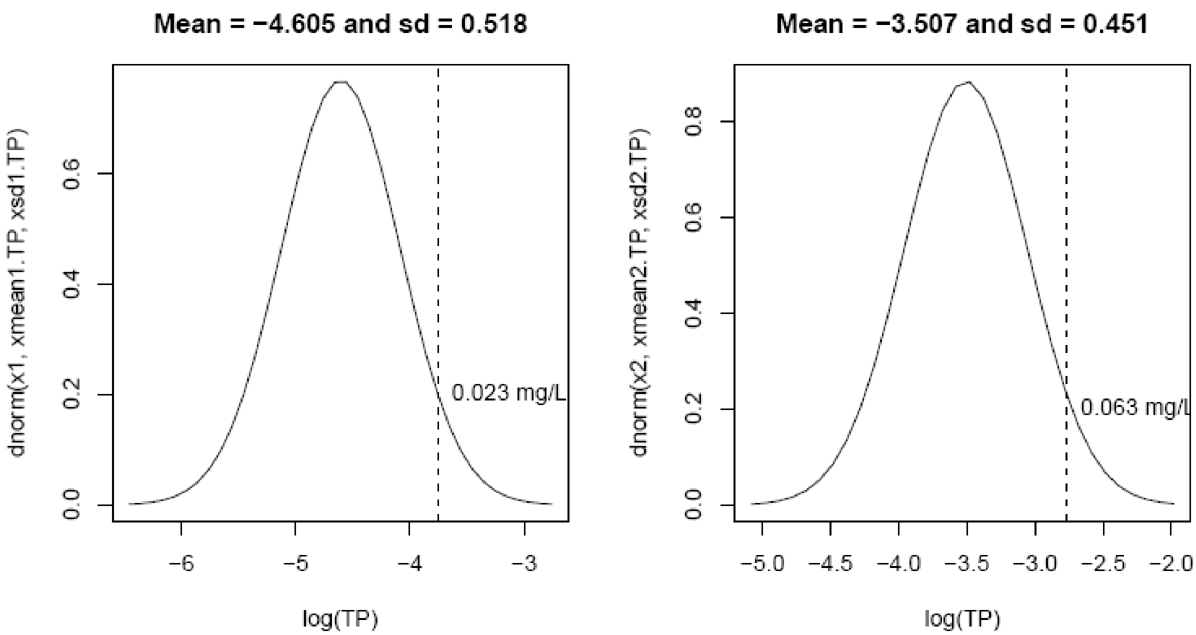


Figure 1-20. Density distribution function of normal distribution for acidic (left) and alkaline lakes (right)